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# 365 Days of progress in surgical oncology

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# 365 Days of progress in surgical oncology

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# Comparison of clinical safety and feasibility between reduced-port laparoscopic radical gastrectomy and conventional laparoscopic radical gastrectomy: A retrospective study

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**Background:** Traditional open gastric cancer surgery has evolved from porous to reduced-hole, single-hole, or even natural cavity surgery to laparoscopic surgery, due to the continuous development of minimally invasive concepts and medical technologies, as well as awareness for the concept of rapid recovery. Conventional laparoscopic radical gastrectomy is quite mature in age at the moment, but how to progress to minimally invasive surgery without increasing the difficulty of surgery while ensuring clinical safety and feasibility is worth further investigation. Therefore, the clinical safety and feasibility of reduced port laparoscopic radical gastrectomy were assessed in this study.
**Methods:** Information on the clinical data of patients undergoing laparoscopic radical gastric cancer surgery in a single centre between May 2020 and May 2022 was collected, and a total of 232 patients were included in this study according to the study protocol design. The clinical data of 232 patients with gastric cancer treated by two different surgical methods, namely, reduced port laparoscopic surgery (RPLS) or conventional laparoscopic surgery (CLS), were retrospectively analysed. The intraoperative indices, postoperative pathological indices, and short-term postoperative complications (within 30 days) of the two different surgical methods were evaluated, as well as the surgical methods' feasibility and short-term postoperative recovery effect.
**Results:** There was no significant difference between the general data of patients with RPLS and CLS ($P > 0.05$). Compared with CLSG, the operation time, digestive tract reconstruction time and lymph node dissection time of RPLSG are shorter. The intraoperative blood loss was less, and the incision was minimally invasive ($P < 0.05$). In the short-term postoperative effect, the level of white blood cell count on the first day, the time of getting out of bed, the time of removing drainage tube, the time of hospitalization and the VAS of pain on the first, third and fifth days after operation, RPLSG was obviously superior to CLSG ($P < 0.05$). There was no significant difference between

Abbreviations
ASA, American Society of Anaesthesiologists; CLS, conventional laparoscopic surgery; RPLS, reduced port laparoscopic surgery; SILS, single-incision laparoscopic surgery; NOSES, Natural orifice specimen extraction surgery; SSI, Surgical-site infection.

RPLSG and CLSG in terms of pathological indices ($P > 0.05$).

**Conclusions:** The treatment of gastric cancer with RPLS has good safety, feasibility and short-term postoperative effects, which is in line with the implementation of the modern concept of rapid rehabilitation surgery.

KEYWORDS

conventional laparoscopic surgery, reduced port laparoscopic surgery, single-incision laparoscopic surgery, natural orifice specimen extraction surgery, gastric cancer

# Introduction

The laparoscopic technique has been gradually utilized in the surgical treatment of early gastric cancer since the application of laparoscopic-assisted radical resection of regional gastric cancer was first reported in 1994 by Kitano et al. (1). Research results of JLSSG-0901 (2) in Japan, KLASS-02 (3) in South Korea and Class-01 (4) in China indicated that laparoscopic radical gastrectomy for locally advanced gastric cancer by professional surgeons did not increase major surgical complications (5, 6). Laparoscopic magnification technology not only enables viewing of fine structures in the vascular system, nerve and fascia in detail, but with the development of endoscopic technology, this further allows the operator to have a special advantage in the clear identification of each anatomical level during the operation. Compared with traditional open surgery, laparoscopic surgery is associated with less pain, less blood loss, a more beautiful incision, fewer inflammatory reactions, faster recovery of gastrointestinal function and shorter hospital stays (7). A consensus, it is widely used in surgical treatment. Conventional laparoscopic surgery (CLS) is mostly conducted by the 5-port method. However, single-incision laparoscopic surgery (8) (SILS) is a single incision of approximately 4 cm (9) through the natural folds of the umbilical region that is placed in a single-port operating platform. The operation is completed through multiple channels on the platform, and it is mostly used for gallbladder and appendix operations (10, 11). Reduced port laparoscopic surgery (RPLS), on the other hand, is based on a single incision through the navel, similar to SILS, and a 12 mm trocar hole is added to the left upper abdomen, through which the abdominal drainage tube can be placed after surgery. The clinical data of 232 gastric cancer patients who met the research plan were retrospectively compared in this study, and the clinical safety and feasibility of laparoscopic radical gastrectomy with a reduced port were assessed.

# Materials and methods

## Patients

Information on the clinical data of patients undergoing laparoscopic radical gastric cancer surgery in a single centre between May 2020 and May 2022 was collected, and a total of 232 patients were included in this study according to the study protocol design, with 176 male patients and 56 female patients and an average age of $57.57 \pm 10.04$ years. They were divided into two groups: CLS ($n = 116$) and RPLS ($n = 116$). The Ethics Committee of Qinghai University's Affiliated Hospital approved the study (approval letter ethics batch number: *P*-SL-20190003), and the patients and their families signed an informed consent form. All of the operations were performed by the same surgical team.

## Inclusion and exclusion criteria

Inclusion criteria were: (1) Age 18–80 years; (2) Before operation, diagnosis was confirmed by pathological biopsy with an ultrasonic gastroscope, and the location and clinical stage of the lesion were further confirmed by contrast-enhanced CT examination of the stomach; (3) Preoperative imaging examination excluded distant metastasis to the liver, lung and other organs; (4) The pathological diagnosis after laparoscopic radical gastrectomy was R0 resection; and (5) Complete clinical data.

Exclusion criteria were: (1) Stage T4b tumour, preoperative existence of fusion lymph nodes, or distant metastasis of tumour; (2) Emergency surgical treatment for complications such as gastric bleeding and perforation before operation; (3) Palliative treatment or conversion to laparotomy during operation; (4) Neoadjuvant chemotherapy before operation; and (5) Incomplete clinical data.

## Operation method and postoperative treatment

The operation methods and postoperative treatment measures were explained to the patients in detail before the operation. According to the patients' wishes, the CLSG (conventional laparoscopic surgery group) or the RPLSG (reduced port laparoscopic surgery group) was freely chosen, and the consent form was signed for the selected operation. The scope of gastric resection and lymph node dissection were all implemented according to the provisions of the «Fifth Edition of Japanese Gastric Cancer Treatment Guidelines» (10). The CLS is laid out

in the conventional five-port method, with a 1-cm-long arcing port along the inferior border of the umbilicus. A 12 mm trocar and a 5 mm trocar were placed 2 cm below the intersection of the anterior axillary line and the rib arch on each side. A 10 mm trocar and a 5 mm trocar were placed at the intersection of the horizontal Line 2 cm above the umbilicus and the lateral border of the rectus abdominis muscle. For RPLS, a 3–5 cm long curved incision was made around the umbilicus at the natural fold of the umbilicus, and a single-port operating platform was placed into the abdominal cavity layer by layer. A 12-mm trocar was then placed 2 cm below the intersection of the patient's left midclavicular line and rib margin. The layout of the surgical puncture port in both groups is shown in **Figures 1A,B**. The patient was placed in a supine split-legged position intraoperatively, as shown in **Figure 1C**. The postoperative abdominal wall incision of the RPLSG patient is shown in **Figure 1D**. For CLS operator position: The main knife is located on the left side or between the legs of the patient, the first aid is located on the right side of the patient, and the laparoscopic assistant is located between the legs or on the right side of the patient. For RPLS operator's position: The main knife is located between the legs of the patient, and the laparoscopic assistant stands on the right side of the patient.

## Observation index

**General information:** sex, age, body mass index, American Society of Anaesthesiologists (ASA) grade, previous abdominal surgery history, tumour length and diameter, tumour location, and tumour differentiation degree;

**Intraoperative indicators:** operation time, digestive tract reconstruction time, lymph node dissection time, intraoperative blood loss, and total length of abdominal incision;

**Postoperative pathological indices:** the total number of lymph nodes, the positive number of metastatic lymph nodes, the distance of the oral margin, the distance of the anal margin, and pT stage, pN stage and pTNM stage.

**Postoperative recovery:** laboratory test indices, postoperative time to getting out of bed, postoperative exhaust time, postoperative intake of liquid diet time, drainage tube removal time, postoperative hospitalization time, visual analogue scale (VAS) on the 1st, 3rd and 5th postoperative days;

**Postoperative safety indicators:** Complications include anastomotic leakage, anastomotic bleeding, pulmonary infection, incision-related complications and pancreatic fistula (Clavien–Dindo Grades II and III) (12).

## Statistical analysis

SPSS 25.0 statistical software was used to analyse the data. When the measurement data were in accordance with the normal distribution, the $t$ or $t'$ test of two independent samples was used and expressed by $(X \pm S)$; when it did not conform to the normal distribution, the rank sum test was used and expressed by $M\ (Q_L - Q_U)$. The qualitative data were tested by the $X^2$ test. When $P < 0.05$, the difference was considered statistically significant. GraphPad Prism 7.00 software was used for statistical graphs.

# Results

## Preoperative general information

According to the research plan, 232 patients with gastric cancer were included in this study, with 116 patients in the CLSG, including 86 males (74.13%) and 36 females (25.87%), with an average age of 56.76 ± 9.37 years old. In addition, 116 patients were in the RPLSG, including 90 males (77.58%) and 26 females (22.42%). The average age for the RPLSG was 58.39 ± 10.65 years old. Statistical analysis showed that there was no significant difference in sex ratio or age between the two groups. Moreover, there was no significant difference between the two groups in BMI (body mass index), ASA (American Society of Anaesthesiologists score) grade, history of previous abdominal surgery, tumour major axes, tumour minor axes, tumour location, or degree of differentiation (see **Table 1**).

## The time of RPLS is shorter, the amount of blood loss is less, and the incision is less invasive

No patients were converted to laparotomy after undergoing laparoscopic radical gastrectomy for gastric cancer in either group. The operation was completed successfully by all 116 RPLS patients, with no additional puncture holes required. The comparison of intraoperative indices between the two groups showed that RPLSG was shorter than CLSG in operation time (**Figure 2A**), digestive tract reconstruction time and lymph node dissection time ($P < 0.05$). Compared with CLSG in intraoperative blood loss and total length of abdominal incision (all trocar puncture sites and auxiliary incisions are included), RPLS was significantly more minimally invasive ($P < 0.05$) (**Figures 2B,C**) (see **Table 2**).

## RPLS can achieve the same radical effect as CLS

In terms of postoperative pathological indices of the two groups, we found that there was no statistical significance in the total number of lymph nodes obtained, positive number

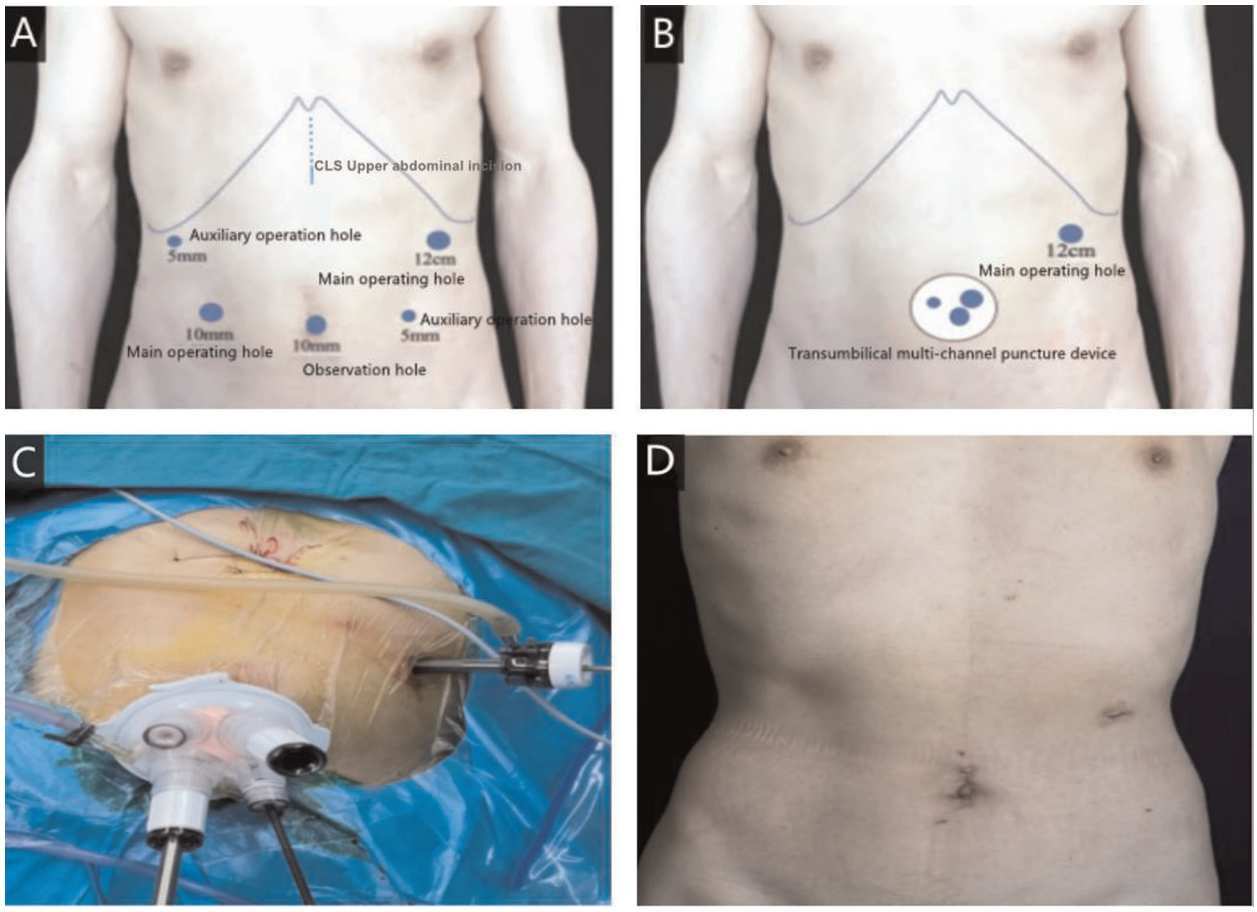


FIGURE 1
(A) the layout of the surgical puncture port of CLS; (B) the layout of the surgical puncture port of RPLS; (C) intraoperative incision position of RPLS; (D) postoperative abdominal wall incision after RPLS.

of metastatic lymph nodes, distance of oral margin, distance of anal margin, pT stage, pN stage or pTNM stage ($P > 0.05$) (See Table 3).

## RPLS can reduce postoperative inflammatory reactions and pain and can accelerate the postoperative recovery of patients

In terms of postoperative recovery, there were statistically significant differences between the two groups in the levels of white blood cell count measured on the first day, albumin measured on the third day, postoperative bed time, postoperative exhaust time, postoperative feeding time, drainage tube removal time, postoperative hospitalization time and VAS score at one day, three days, and five days after operation ($P < 0.05$) (Figures 3A–C). However, there was no significant difference in white blood cell count, haemoglobin or total bilirubin on the third and fifth days ($P > 0.05$) (See Tables 4, 5).

## RPLS has the same security as CLS and can reduce the occurrence of SSI

In terms of postoperative safety indicators, there was no significant difference in the incidence of anastomotic leakage, anastomotic bleeding or pulmonary infection between the two groups ($P > 0.05$), but there were significant differences in the incidence of incision-related complications and pancreatic fistula ($P < 0.05$) (Figure 4) (See Table 6).

# Discussion

With the increasing development of minimally invasive and standardized surgery, laparoscopic surgery has evolved from

TABLE 1 Preoperative patient demographic information.

| | CLSG (n = 116) | RPLSG (n = 116) | *P*-value |
|---|---|---|---|
| Age (years) | 56.76 ± 9.37 | 58.39 ± 10.65 | 0.220 |
| Gender (%) | | | |
| Male | 86 (74.13%) | 90 (77.58%) | 0.539 |
| Female | 30 (25.87%) | 26 (22.42%) | |
| BMI (kg/m$^2$) | 23.11 (20.20–25.58) | 22.06 (20.00–24.42) | 0.067 |
| ASA grade (%) | | | |
| I | 4 (3.44%) | 9 (7.75%) | 0.194 |
| II | 85 (73.27%) | 74(63.79%) | |
| III | 27 (23.29%) | 33 (28.46%) | |
| History of previous abdominal surgery (%) | | | |
| No | 70 (60.34%) | 83 (71.55%) | 0.096 |
| Yes | 46 (39.66%) | 33 (28.45%) | |
| Tumour major axes (cm) | 3.50 (3.00–4.00) | 3.00 (2.52–3.90) | 0.066 |
| Tumour minor axes (cm) | 3.00 (2.00–3.00) | 2.40 (1.90–3.00) | 0.073 |
| Tumour location (%) | | | |
| Upper 1/3 of the stomach | 36 (31.03%) | 32 (27.58%) | 0.221 |
| Middle 1/3 of the stomach | 28 (24.13%) | 40(34.48%) | |
| Lower 1/3 of the stomach | 52 (44.84%) | 44 (37.94%) | |
| Degree of differentiation (%) | | | |
| Highly differentiated | 10 (8.62%) | 5 (4.31%) | 0.270 |
| Intermediate differentiation | 32 (27.58%) | 40 (34.48%) | |
| Low differentiation | 74 (63.80%) | 71 (61.21%) | |

TABLE 2 Intraoperative correlation index.

| | CLSG (*n* = 116) | RPLSG (*n* = 116) | *P*-value |
|---|---|---|---|
| Operation Time (min) | 275 (240–300) | 240 (210–280) | 0.002 |
| Digestive tract reconstruction time (min) | 80 (70–100) | 70 (60–80) | 0.001 |
| Lymph node dissection time (min) | 200 (170–220) | 170 (140–190) | 0.001 |
| Intraoperative bleeding volume (ml) | 50 (30–50) | 10 (10–50) | 0.001 |
| Total length of abdominal incision (cm) | 8.60 (8.40–8.90) | 5.40 (5.20–5.60) | 0.001 |

multiport to reduced-port, single-port, and even natural orifice specimen extraction surgery (NOSES) (13). The progression of minimally invasive surgery technology is the result of the combined advancement of surgical concepts, surgical instruments, and surgical techniques. The aim of minimally invasive surgery is to provide a painless and scar-less surgical approach (14). In terms of cosmetology and accelerated rehabilitation surgery, a large number of literature reports (15, 16) show that laparoscopic radical gastrectomy is superior to open surgery. Traditional laparoscopic radical gastrectomy for gastric cancer uses a five-port method with or without liver suspension, and gastric dissociation, lymph node dissection, and digestive tract reconstruction are completed with the help of assistants. Omori et al. (15) were the first to report the use of single-port laparoscopic radical gastrectomy for early distal gastric cancer in 2011. However, because of the lack of triangular positioning between the surgical instruments and the abdominal lens, coaxial effects easily occur, which limits the operating range of the surgical area and causes rear-end collisions between surgical instruments—not only increasing the operation difficulty but also placing higher demands on the supporting surgical team (17). Simultaneously, more clinical trials are required to confirm the curative effect of oncology, lymph node dissection, and digestive tract reconstruction. As a result, single-port laparoscopic radical gastrectomy development is limited, and it is more frequently used in simple operations such as cholecystectomy and appendectomy (18–21). In contrast, RPLS uses an additional 12-mm poking port 2 cm below the intersection of the left

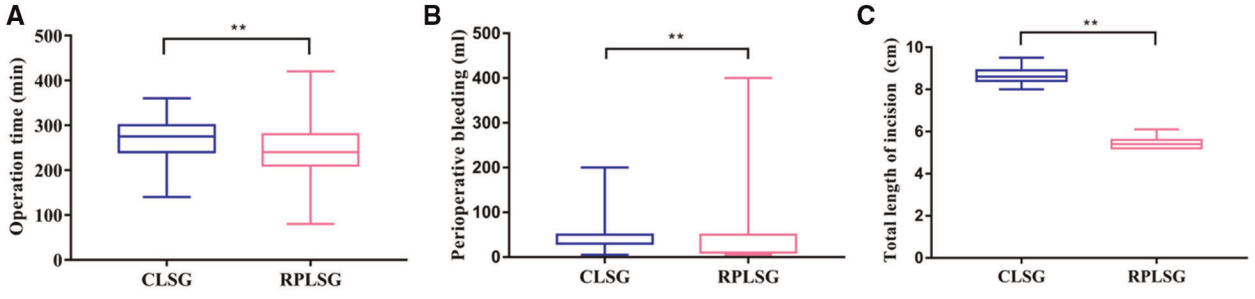


FIGURE 2
(A) operation time of different surgical methods; (B) effect of different surgical methods on perioperative bleeding; (C) effect of different surgical methods on total length of incision.

midclavicular line and the rib margin as the main operating port of the main knife minus the two holes of the assistant, based on the SILS. This method can facilitate the clearance of regional lymph nodes in the suprapancreatic region and the splenic hilar region, while overcoming the operational drawbacks associated with single-port laparoscopy. At the same time, it can be used to place the abdominal drainage tube without making another abdominal incision, which reduces damage to the abdominal wall blood vessels and nerves, not only improving surgical safety but also balancing the relationship between surgical safety and minimally invasive surgery. As a result, some surgeons will attempt to use RPLS with laparoscopic assistance to complete gastric dissociation, lymph node dissection, and digestive tract reconstruction.

Although CLS is less difficult than RPLS and should take less time in operation, digestive tract reconstruction, and lymph node dissection, the results of this study show that RPLS takes less time in operation, digestive tract reconstruction, and lymph node dissection, contradicting conventional knowledge. The absence of the trocar incision in RPLS may lead to an increase in the difficulty of the procedure and a prolongation in time of the procedure. When an RPLS surgeon has completed the RPLS learning curve and their surgical technique and proficiency have improved, the precision of intraoperative operations will be increased. At the same time, the surgeon can complete gastric dissociation, lymph node dissection, and digestive tract reconstruction with the help of a laparoscopic assistant, and the coordination of one person's actions is better than that of the assistant's, which is one of the main reasons for shortening the operation time, digestive tract reconstruction time, and lymph

TABLE 3 Postoperative pathological indices.

| | CLSG ($n = 116$) | RPLSG ($n = 116$) | *P*-value |
|---|---|---|---|
| Total number of lymph nodes obtained | 34.43 ± 15.07 | 35.06 ± 13.03 | 0.734 |
| Number of positive lymph node metastases | 1.00 (0.00–7.00) | 1.00 (0.00–7.00) | 0.949 |
| Mouth-side margin distance (cm) | 2.50 (1.50–4.00) | 2.50 (1.00–4.50) | 0.394 |
| Anal margin distance (cm) | 3.75 (2.00–6.00) | 3.65 (2.00–6.00) | 0.728 |
| Staging of pT (%) | | | |
| $pT_1$ stage | 16 (13.79%) | 20 (17.24%) | 0.443 |
| $PT_2$ stage | 26 (22.41%) | 23 (19.82%) | |
| $PT_3$ stage | 51 (43.96%) | 42 (36.20%) | |
| $PT_4$ stage | 23 (19.84%) | 31 (26.74%) | |
| Staging of pN (%) | | | |
| $pN_0$ stage | 53 (45.68%) | 50 (43.10%) | 0.172 |
| $pN_1$ stage | 13 (11.20%) | 25 (21.55%) | |
| $pN_2$ stage | 17 (14.65%) | 12 (10.34%) | |
| $pN_3$ stage | 33 (28.47%) | 29 (25.01%) | |
| Staging of pTNM (%) | | | |
| I stage | 33 (28.44%) | 28 (24.13%) | 0.684 |
| II stage | 32 (27.58%) | 31 (26.72%) | |
| III stage | 51 (43.98%) | 57 (49.15%) | |

TABLE 4 Postoperative recovery index.

| | CLSG ($n = 116$) | RPLSG ($n = 116$) | *P*-value |
|---|---|---|---|
| Postoperative bedtime (h) | 48.00 (24.00–48.00) | 24.00 (24.00–24.00) | 0.001 |
| Postoperative time to exhaustion (h) | 72.00 (48.00–72.00) | 48.00 (48.00–72.00) | 0.001 |
| Postoperative feeding time (d) | 9.00 (7.00–10.00) | 8.00 (6.00–9.00) | 0.002 |
| Drainage tube removal time (d) | 10.00 (8.00–12.00) | 8.00 (5.00–11.00) | 0.001 |
| Postoperative hospitalization time (d) | 11.50 (9.00–14.00) | 11.00 (8.00–13.00) | 0.007 |
| VAS | | | |
| Day 1 | 4.59 ± 0.80 | 3.52 ± 0.95 | 0.001 |
| Day 3 | 3.45 ± 0.77 | 2.46 ± 0.87 | 0.001 |
| Day 5 | 2.72 ± 1.16 | 1.63 ± 0.72 | 0.001 |

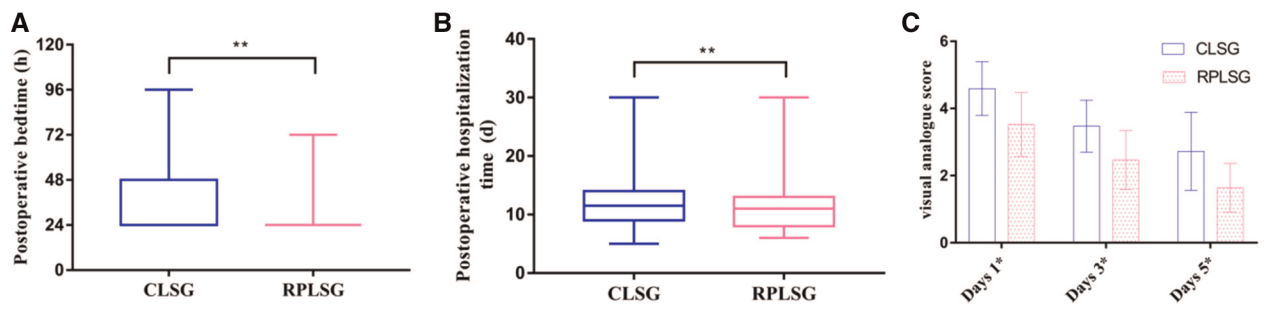


FIGURE 3
(A) effect of different surgical methods on postoperative bedtime; (B) effect of different surgical methods on postoperative hospitalization time; (C) the effect of different surgical methods on postoperative pain on days 1, 3, and 5.

TABLE 5 Postoperative recovery index.

| | CLSG ($n = 116$) | RPLSG ($n = 116$) | *P*-value |
|---|---|---|---|
| White blood cell count ($10^9$/L) | | | |
| Day 1 | 11.39 (9.86–14.16) | 10.67 (8.79–12.60) | 0.014 |
| Day 3 | 8.37 (6.12–10.40) | 7.89 (6.07–9.90) | 0.291 |
| Day 5 | 6.13 (4.92–8.25) | 6.04 (5.01–7.87) | 0.841 |
| Albumin level (g/L) | | | |
| Day 1 | 35.60 (33.30–38.37) | 36.70 (33.82–38.80) | 0.062 |
| Day 3 | 36.55 (34.05–38.07) | 36.95 (35.00–39.72) | 0.005 |
| Day 5 | 36.90 (34.55–40.15) | 38.00 (35.62–40.57) | 0.112 |
| Haemoglobin level (g/L) | | | |
| Day 1 | 129.96 ± 26.33 | 127.38 ± 24.65 | 0.442 |
| Day 3 | 115.07 ± 21.68 | 116.43 ± 21.55 | 0.634 |
| Day 5 | 113.32±21.35 | 114.26±21.53 | 0.739 |
| Total bilirubin level (μmol/L) | | | |
| Day 1 | 13.80 (9.52–19.62) | 14.70 (9.82–22.80) | 0.365 |
| Day 3 | 17.80 (14.00–25.07) | 18.10 (13.12–25.57) | 0.900 |
| Day 5 | 20.10 (14.17–29.30) | 17.85 (14.25–29.30) | 0.472 |

TABLE 6 Postoperative safety index.

| | CLSG ($n = 116$) | RPLSG ($n = 116$) | *P*-value |
|---|---|---|---|
| Anastomotic fistula (%) | | | |
| No | 104 (89.65%) | 110 (94.82%) | 0.219 |
| Yes | 12 (10.35%) | 6 (5.18%) | |
| Anastomotic bleeding (%) | | | |
| No | 114 (98.27%) | 115 (99.13%) | 1.000 |
| Yes | 2 (1.73%) | 1 (0.87%) | |
| Pulmonary infection (%) | | | |
| No | 109 (93.96%) | 113 (97.41%) | 0.333 |
| Yes | 17 (6.04%) | 3 (2.59%) | |
| Incision-related complications (%) | | | |
| No | 101 (87.06%) | 114 (98.27%) | 0.002 |
| Yes | 15 (12.94%) | 2 (1.73%) | |
| Pancreatic fistula (%) | | | |
| No | 105 (90.51%) | 116 (100%) | 0.001 |
| Yes | 11 (9.49%) | 0 (0.00%) | |

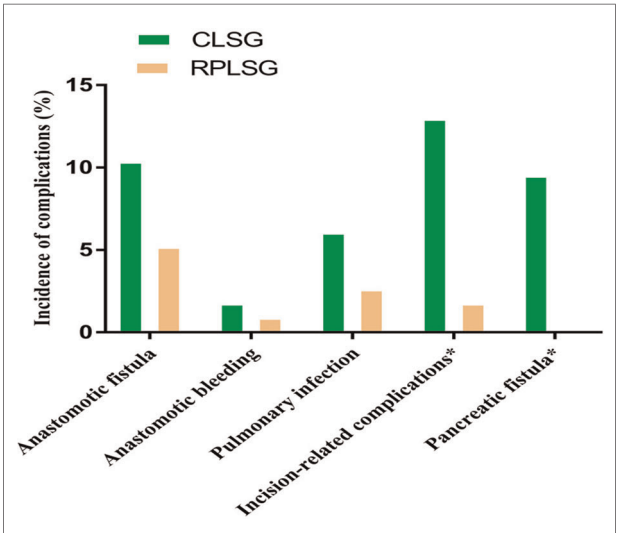


FIGURE 4
Effect of different surgical methods on the incidence of postoperative complications.

node dissection time. Furthermore, in conventional laparoscopic radical gastrectomy for gastric cancer, the assistant frequently causes tissue traction and accessory damage to organs in the operation area due to insufficient cooperation, resulting in a corresponding extension of the operation procedure and an increase in postoperative complications (22–23). RPLS, on the other hand, is operated independently by the chief surgeon, which can avoid issues caused by improper operation team cooperation, thus improving operation efficiency and reducing intraoperative blood loss. The CLS multiple trocar puncture port approach may decrease patient satisfaction with the postoperative aesthetics of the abdominal wall incision. In addition, this approach also increases the risk of complications associated with trocar port herniation, infection, and metastatic tumour cell implantation. After the completion of endoscopic dissociation, CLS requires reselection of the abdominal wall incision to remove the specimen. However, the reselection of the incision will inevitably lead to a longer operative time. It also increases the total length of the abdominal incision because of the increased number of trocar puncture ports, which may lead to an increased incidence of intraoperative and postoperative abdominal infection and surgical-site infection (SSI) (24). This results in increased postoperative pain, delayed incisional healing, reduced abdominal wall aesthetics, and increased financial costs and psychological burden for the patient. In contrast, for RPLS, there is no need to reselect the abdominal wall incision, and the operation is completed by removing the specimen through a single incision in the umbilicus, using the curvature of the umbilicus after pulling out the single-port operating platform. This brings great convenience to the operation. The umbilical incision has natural folds due to the low fatty and muscle tissue content in the abdominal wall layer. Postoperatively, the incision is better concealed than CLS, and the patient has better postoperative abdominal wall aesthetics with less postoperative pain. This also facilitates early postoperative bed and out-of-bed activities and promotes rapid recovery of patient function. As RPLS is less invasive, it can reduce the postoperative inflammatory response and has greater advantages in accelerating postoperative rehabilitation in patients under the condition of a single operation to avoid side injury and shorter operation

time. Simultaneously, under the condition of a single operation, the amount of postoperative exudation is reduced to avoid side injury and shorter operation time, and the time of abdominal cavity extubation is shortened, avoiding the delay of extubation —which is usually caused by an increase in exudation and increases the probability of abdominal cavity and incision infections and the economic and psychological burden on patients. In terms of postoperative safety and pathological indicators, there was no significant difference in the incidence of anastomotic leakage, anastomotic bleeding, pulmonary infection, or the total number of lymph nodes between the two groups, indicating that RPLS can still achieve the same radical effect as CLS without increasing postoperative complications, but the use of reduced-port laparoscopy in radical treatment of gastric cancer still requires a large number of clinical studies for further confirmation.

RPLS technology was developed on the basis of CLS, which avoided the difficulty of SILS operation and served as a bridge between SILS and CLS. However, only when the operator is proficient in CLS and has overcome the RPLS learning curve can the operator complete the operation with the help of a laparoscopic assistant, which not only saves manpower but also prevents intraoperative side injuries and improves operation time and efficiency. Although the multichannel puncture platform used in RPLS will impose some financial burden on patients, the short-term postoperative effect of patients suggests that it is a potentially feasible and inexpensive way to mitigate economic costs after surgery. Of course, we discovered a report that (25) can easily create this type of instrument platform during operation, which is simple to use, economical, and feasible. However, there are some concerns about this operation right now, such as a lack of training assistants and surgical teams. This operation, however, can only be performed after standardized and rational training and mastery of laparoscopic radical gastrectomy for gastric cancer. As a result, the operation continues to emphasize operation team cooperation and assistant training while placing greater emphasis on the operation's skill and safety. As a result, intraoperative side injury is avoided, perioperative complications are reduced, the operation is made less invasive, patients' postoperative rehabilitation is accelerated, and patients benefit. However, the RPLS umbilical incision length limitation is also one of the reference factors for tumour length and diameter selection. The resected tumour focus is bound to be removed from the umbilical single incision. If the tumour focus is too large, it may not be removed, so it is necessary to further extend the umbilical incision, which not only increases the trauma of RPLS but also prolongs the operation time and increases the probability of incision infection in the operation area. Therefore, the tumour length and diameter of all patients in this study were $\leq$4 cm to ensure the smooth removal of the tumour focus through the umbilical single incision.

## Conclusions

Our findings show that laparoscopic radical gastrectomy with reduced-port laparoscopy is clinically safe and feasible. Compared to CLS, it has the advantages of less trauma, fewer inflammatory reactions, and better cosmetic effects, which can accelerate patients' postoperative recovery and is more in line with the modern concept of rapid rehabilitation surgery and minimally invasive surgery.

Although the data from this study can be used to support clinical surgeons to perform this procedure, the sample size is small and based on a single-centre data study. More research samples are required to confirm the feasibility and safety of RPLS and to clarify the surgical application value of this method.

## Data availability statement

The raw data supporting the conclusions of this article will be made available by the authors, without undue reservation.

## Ethics statement

The study was approved by the Ethics Committee of the Qinghai University Affiliated Hospital, and all patients signed informed consent forms (approval number P-SL-20190003).

## Author contributions

XC contributed to the study concept and design. SY and XM conducted the laparoscopic radical gastrectomy. LW and YD wrote the manuscript. CW and WM collected and analyzed the data. XC revised and edited the manuscript. SY and XM are the guarantors of this study. All authors contributed to the article and approved the submitted version.

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## Conflict of interest

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

## Publisher's note



## References

1. Kitano S, Iso Y, Moriyama M, Sugimachi K. Laparoscopy-assisted billroth: I gastrectomy. *Surg Laparosc Endosc*. (1994) 4(2):146–8. doi: 10.1097/sle.0000000000000011

2. Inaki N, Etoh T, Ohyama T, Uchiyama K, Katada N, Koeda K, et al. A multi-institutional, prospective, phase II feasibility study of laparoscopy-assisted distal gastrectomy with D2 lymph node dissection for locally advanced gastric cancer (JLSSG0901). *World J Surg*. (2015) 39(11):2734–41. doi: 10.1007/s00268-015-3160-z

3. Hyung WJ, Yang HK, Park YK, Lee HJ, An JY, Kim W, et al. Long-term outcomes of laparoscopic distal gastrectomy for locally advanced gastric cancer: the KLASS-02-RCT, randomized clinical trial. *J Clin Oncol*. (2020) 38 (28):3304–13. doi: 10.1200/JCO.20.01210

4. Yu J, Huang C, Sun Y, Su X, Cao H, Hu J, et al. Effect of laparoscopic vs open distal gastrectomy on 3-year disease-free survival in patients with locally advanced gastric cancer: the CLASS-01 R- andomized clinical trial. *JAMA*. (2019) 321 (20):1983–92. doi: 10.1001/jama.2019.5359

5. Kim HI, Hur H, Kim YN, Lee HJ, Kim MC, Han SU, et al. Standardization of D2 lymphadenectomy and surgical quality control(KLASS-02-QC):a prospective, observational,multicenter study [NCT01283893]. *BMC Cancer*. (2014) 14:209. doi: 10.1186/1471-2407-14-209

6. Hu Y, Huang C, Su Y, Su X, Cao H, Hu J, et al. Morbidity and mortality of laparoscopic versus open D2 distal gastrectomy for advanced gastric cancer: a randomized controlled trial. *J Clin Oncol*. (2016) 34(12):1350–7. doi: 10.1200/JCO.2015.63.7215

7. Wang H, Zhao X, LI G, FU Y, Kuang L, Cui S, et al. Safety and short-term efficacy of laparoscopic assisted distal gastrectomy versus open distal gastrectomy in D2 radical surgeries for locally advanced distal gastric cancer: a meta-analysis. *J China Med Univ*. (2015) 3(44):252–8. doi: 10.3969/j.issn.0258-4646.2015.03.014

8. Ma B, Zhou J, Li JG, Wang J. Safety evaluation of single-port +1-port laparoscopic radical resection of distal gastric cancer. *Chin J Gen Surg*. (2021) 15(6):653–6. doi: 10.3877/cma.j.issn.1674-3946.2021.06.019

9. Cui W, Li T, Li S. 20 Years of laparoscopic gastric cancer surgery and achievements in China. *Chin J Gen Surg*. (2021) 15(2):139–42. doi: 10.3877/cma.j.issn.1674-3946.2021.02.005

10. Ito E, Takai A, Imai Y, Otani H, Onishi Y, Yamamoto Y, et al. Quality of life after single-incision laparoscopic cholecystectomy: a randomized, clinical trial. *Surgery*. (2019) 165(2):353–9. doi: 10.1016/j.surg.2018.08.004

11. Maggiori L, Tuech JJ, Cotte E, Lelong B, Denost Q, Karoui M, et al. Single-incision laparoscopy versus multiport laparoscopy for colonic surgery: a multi center, double-blinded, randomized controlled trial. *Ann Surg*. (2018) 268(5):740–6. doi: 10.1097/SLA.0000000000002836

12. Japanese Gastric Cancer Association. Japanese Gastric cancer treatment guidelines 2018 (5th edition). *Gastric Cancer*. (2021) 24(1):1–21. doi: 10.1007/s10120-020-01042-y

13. Hirano Y, Hiranuma C, Hattori M, Douden K, Yamaguchi S. Single-incision or single-incision plus one-port laparoscopic surgery for colorectal cancer. *Surg Technol Int*. (2020) 36:132–5. doi: 10.1186/s13063-015-1067-5

14. Zheng MH, Ma JJ. Concept innovation: a new perspective on minimally invasive surgery . *Chin J Gastrointest Surg*. (2020) 19(5):478–81. doi: 10.3760/cma.j.cn115610-20200413-00256

15. Yan Q, Ma XF, Zhao K, Chen XQ, Guo C, Wang QQ, et al. Analysis of technical difficulties of single-port and reduced-port laparoscopic radical gastric cancer surgery. *Chin J Gastrointest Surg*. (2019) 18(3):222–8. doi: 10.3760/cma.j.issn.1673-9752.2019.03.006

16. Morales-Conde S, Peeters A, Meyer YM, Antoniou SA, Del Agua IA, Arezzo A, et al. European Association for endoscopic surgery (EAES) consensus statement on single-incision endoscopic surgery. *Surg Endosc*. (2019) 33 (4):996–1019. doi: 10.1007/s00464-019-06693-2

17. Jin P, Tian YT. Advances and controversies in the treatment of single-port laparoscopic radical gastric cancer. *J Laparoscopic Surg*. (2020) 25(1):1–3. doi: CNKI:SUN:FQJW.0.2020-01-001

18. Lee Y, Kim HH. Single-incision laparoscopic gastrectomy for gastric cancer. *J Gastric Cancer*. (2017) 17(3):193–203. doi: 10.5230/jgc.2017.17.e29

19. Katsuyama S, Nakajima K, Kurokawa Y, Takahashi T, Miyazaki Y, Makino T, et al. Single-incision laparoscopic intragastric surgery for gastric submucosal tumor located adjacent to esophagogastric junction: report of four cases. *J Laparoendosc Adv Surg Tech A*. (2018) 28(1):78–82. doi: 10.1089/lap.2017.0026

20. Tei M, Otsuka M, Suzuki Y, Kishi K, Tanemura M, Akamatsu H. Safety and feasibility of single-port laparoscopic multivisceral resection for locally advanced left colon cancer. *Oncol Lett*. (2018) 15(6):10091–7. doi: 10.3892/ol.2018.8582

21. Liu X, Li JB, Shi G, Guo R, Zhang R. Systematic review of single-incision versus conventional multiport laparoscopic surgery for sigmoid colon and rectal cancer. *World J Surg Oncol*. (2018) 16(1):220. doi: 10.1186/s12957-018-1521-4

22. Lee SH, Kim KH, Choi CW, Kim SJ, Kim DH, Choi CI, et al. Atraumatic liver retraction using nelaton catheters during totally laparoscopic gastrectomy. *Surg Laparosc Endosc Percutan Tech*. (2017) 27(6):485–90. doi: 10.1097/SLE.0000000000000489

23. Ida S, Hiki N, Ishizawa T, Kuriki Y, Kamiya M, Urano Y, et al. Pancreatic compression during lymph node dissection in laparoscopic gastrectomy: possibleCause of pancreatic leakage. *J Gastric Cancer*. (2018) 18(2):134–41. doi: 10.5230/jgc.2018.18.e15

24. Dusch N, Goranova D, Herrle F, Niedergethmann M, Kienle P. Randomized controlled trial: comparison of two surgical techniques for closing the wound following ileostomy closure: purse stringvsdirect suture. *Colorectal Dis*. (2013) 15(8):1033–40. doi: 10.1111/codi.12211

25. Zhang H, Ling YZ, Cong JC, Cui MM, Liu DS, Chen CS. Comparison of short-term efficacy of modified two-hole approach and conventional five-hole approach for laparoscopic anterior rectal cancer resection . *Chin J Pract Surg*. (2016) 36(10):1084–9. doi: CNKI:SUN:ZGWK.0.2016-10-023

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# The role of surgery in older patients with T1-2N0M0 small cell lung cancer: A propensity score matching analysis

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**Background:** Surgical resection could improve the survival of patients with early-stage small cell lung cancer (SCLC). However, there is a lack of dedicated studies concentrating on surgical treatment in older patients with T1-2N0M0 SCLC. Thus, we performed this population-based study to investigate whether older patients with T1-2N0M0 SCLC could benefit from surgery.

**Methods:** We collected the data of patients with SCLC between 2000 and 2015 from the Surveillance, Epidemiology, and End Results Program database. Older patients (≥ 65 years) with T1-2N0M0 SCLC were included, and we converted the staging information into those of the eighth edition. The propensity score matching (PSM) was used to balance the distribution of clinical characteristics between surgery and no-surgery groups.

**Results:** Before PSM, the distribution proportions of clinical characteristics in 1,229 patients were unbalanced. The Kaplan–Meier curves of overall survival (OS) and cancer-specific survival (CSS) showed that the patients in the surgery group were better than those in the non-surgery group (all $P < 0.001$). After 1:2 PSM, the distribution proportions of clinical characteristics in 683 patients were balanced (all $P > 0.05$). The OS and CSS of patients in the surgery group were still better than that of patients in the no-surgery group (all $P < 0.001$), and subgroup analysis showed that the surgery was a protective factor for OS and CSS in all clinical characteristics subgroups (almost $P < 0.001$). The multivariate Cox analysis further confirmed this result (OS: HR, 0.33; 95% CI, 0.27–0.39; $P < 0.001$; CSS: HR, 0.29; 95% CI, 0.23–0.36; $P < 0.001$). The result of subgroup analysis based on age, T stage, and adjuvant therapy showed that surgery was related to better OS and CSS compared with non-surgery group (almost $P < 0.001$) and that lobectomy exhibited the longer survival than sublobectomy. Age, sex, and race were the independent prognostic factors for OS in patients undergoing surgery, whereas only the factor of age affects the CSS in patients with surgery.

**Conclusions:** Older patients with T1-2N0M0 SCLC can benefit significantly from surgical treatment, and lobectomy provides better prognosis than sublobectomy.

KEYWORDS

small cell lung cancer, older patients, prognosis, propensity score matching, surgery

## Introduction

Lung cancer is the leading cause of malignancy incidence and mortality (1). Small cell lung cancer (SCLC) accounts for approximately 15% of total lung cancer cases and is characterized by rapid growth, high vascularity, early metastatic spread, significant sensitivity to chemotherapy and radiotherapy, and development of drug resistance during the course of disease, with a 5-year survival rate of 7% (2, 3). Thus, stage T1-2N0M0 SCLC only accounts for nearly 5% of patients diagnosed with SCLC, which have a better prognosis, with a 5-year survival up to 50% (4, 5). As the aged population increases, the diagnosis of cancer will continue to rise in older patients. Lung cancer has become a disease of the elderly, with the average age at diagnosis of 70 years (6, 7). The standard treatment for SCLC is chemotherapy alone or in combination with concurrent radiotherapy (8), but the rate of local recurrence is up to 50% in limited stage, although SCLC is sensitive to chemotherapy and radiotherapy (9, 10). Currently, the National Comprehensive Cancer Network (NCCN) guidelines recommended surgery for patients with clinical T1-2N0M0 SCLC (11). Moreover, some retrospective studies stated that patients with limited SCLC who underwent surgery had an excellent survival and 5-year survival rate of approximately 50% (5, 12). In addition, the previous study in terms of surgical approach showed that the survival of the lobectomy group was better than that of the wedge resection in patients with stage I-IIA SCLC (13, 14).

However, compared with young people, the older patients may be frail with complex underlying diseases, poor performance status, and increased treatment-related complications (15). Therefore, identifying the optimal treatment for older patients with early-stage SCLC is challenging. Previous research showed that comorbidity alone was not the reason to withhold standard therapy in limited SCLC (16). Because of the low enrollment of older patients in cancer randomized clinical trials (RCTs) (17), there was also a lack of evidence-based RCTs that surgery is superior to conservative management in terms of long-term survival benefits in the older population. Meanwhile, surgery and postoperative adjuvant therapy were significantly underused among the older population with early-stage SCLC (4, 13, 18). Consequently, there was still no consensus on whether the older patients (≥ 65 years) with T1-2N0M0 SCLC could benefit from surgery currently.

In this study, we performed strict matching of clinical data between the surgical and non-surgical groups by propensity score matching (PSM), so as to eliminate the confounding effect of clinical characteristics of the two groups. Finally, we evaluated the effect of surgery on the long-term survival in older patients (≥ 65 years) with T1-2N0M0 SCLC based on the Surveillance, Epidemiology, and End Results (SEER) Program database.

## Methods

### Patient selection

The SEER database is an authoritative source for cancer statistics that covers approximately 28% of the US population and contains data on cancer occurrences in 18 areas of the United States. The selected patients diagnosed with SCLC were identified using the SEER * Stat version 8.3.9 (National Cancer Institute, Bethesda, MD, USA). The study cohort consisted of the patients with the International Classification of Disease for Oncology Third Edition (ICD-O-3) morphology codes (8041/3, 8042/3, 8043/3, 8044/3, and 8045/3) and site codes (C34.0, C34.1, C34.2, C34.3, C34.8, and C34.9). The exclusion criteria were as follows: (I) not receiving regular follow-up or no follow-up; (II) patients having at least one prior malignancy; (III) not pathologically confirmed by immunohistochemistry; and (IV) patients with missing information concerning primary tumor size (T), regional lymph node (N), or distant metastasis (M) stage and clinical information. After that, we also set up the including criteria for the patients meeting the above exclusion criteria: aged ≥ 65 years; patients with the eighth edition of American Joint Committee on Cancer (AJCC) staging system, stage T1-2N0M0 (Figure 1).

### Variables

To facilitate data analysis, we converted continuous variables into categorical variables. The extracted clinical information

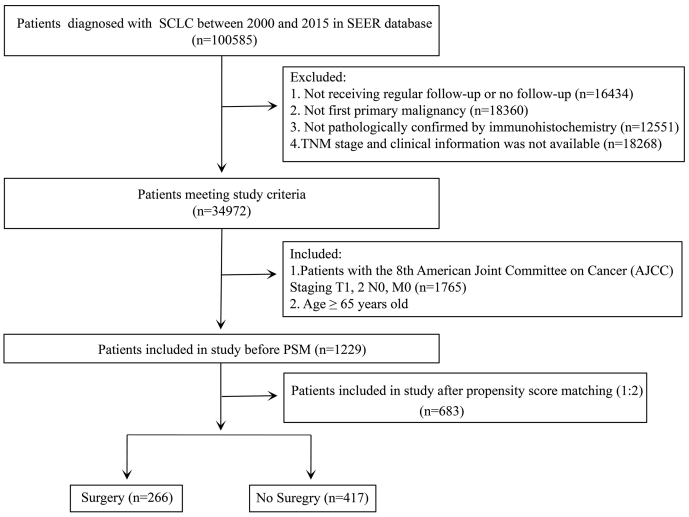


FIGURE 1
Flowchart for data filtration of older patients with T1-2N0M0 small cell lung cancer (SCLC).

included sex, age (65–70, 71–80, and >80 years), race, laterality (left and right), T stage (T1a, T1b, T1c, and T2), surgery (surgery and no surgery), radiotherapy or not, chemotherapy or not, survival months, causes of death, and vital status. In terms of surgery, we defined the resection of less than one lobe as sublobectomy as some surgical procedures were not clear in the SEER database or the number was so small that we cannot analyze them separately. In addition, we converted the TNM categories for each patient according to the Collaborative Staging Manual and Coding Instructions for the eighth edition of the AJCC staging system using tumor size and tumor CS extension. For chemotherapy or radiotherapy, we were unable to define neoadjuvant or adjuvant therapy due to the lack of sequence of the treatment. The primary outcome was defined as overall survival (OS) and cancer-specific survival (CSS). The time of the last follow-up was November 2020. OS was defined as the interval between cancer diagnosis and death resulting from any cause or the last follow-up for patients still alive. CSS was defined as the length of time from cancer diagnosis to death from SCLC.

## Statistical analysis

The baseline characteristics of patients in the surgery group and the no-surgery group are described using frequencies and percentage. PSM was performed between the two groups to reduce potential bias and possible confounding interference. The baseline demographic data for the two groups were compared using the Student's t-test or $\chi^2$ test and the Fisher's exact test before and after PSM as deemed appropriate. Kaplan–Meier survival curves were plotted to assess distinctions in prognosis by applying the log-rank test. We used Cox proportional hazards regression analyses with both univariate and multivariate Cox regression analyses. Moreover, the multivariate Cox proportional hazards regression models were also performed to assess the risk factors in subgroup analyses. In addition, the forest plot of hazard ratios was constructed from univariate and multivariate Cox regression analyses. All data analyses were performed using RStudio version 4.1.2 (RStudio, Boston, MA, USA). A two-sided $P$-value < 0.05 was deemed significant.

# Result

## Baseline clinical characteristics

A total of 1,229 patients aged ≥65 years who had been diagnosed with T1-2N0M0 SCLC were included in our study. Of the population included, 71.6% of patients (880 patients) did not receive the surgical resection. The baseline characteristics of patients and tumors are shown in Table 1. The result showed that the distribution frequencies of some characteristics, including age, race, T stage, radiotherapy, and chemotherapy, were quite unbalanced between the surgery group and the no-surgery group. The patients in the surgery group were fewer than that in the no-surgery group among different age groups. The no-surgery group was associated with the white race and the

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TABLE 1 The clinicopathologic characteristics of older patients with T1-2N0M0 SCLC before propensity score matching.

| Characteristics | Total (N, %) | No Surgery | Surgery | P-value |
|---|---|---|---|---|
| All | 1,229 | 880 (71.60) | 349 (28.4) | |
| Age (year) | | | | <0.001 |
| >80 | 204 (16.60) | 166 (18.86) | 38 (10.89) | |
| 65–70 | 416 (33.85) | 272 (30.91) | 144 (41.26) | |
| 71–80 | 609 (49.55) | 442 (50.23) | 167 (47.85) | |
| Sex | | | | 0.908 |
| Female | 667 (54.27) | 479 (54.43) | 188 (53.87) | |
| Male | 562 (45.73) | 401 (45.57) | 161 (46.13) | |
| Race | | | | 0.004 |
| Black | 94 (7.65) | 79 (8.98) | 15 (4.30) | |
| Other | 50 (4.07) | 41 (4.66) | 9 (2.58) | |
| White | 1085 (88.28) | 760 (86.36) | 325 (93.12) | |
| Laterality | | | | 0.434 |
| Left | 532 (43.29) | 388 (44.09) | 144 (41.26) | |
| Right | 695 (56.55) | 490 (55.68) | 205 (58.74) | |
| Unknown | 2 (0.16) | 2 (0.23) | 0 (0.00) | |
| T stage (eighth edition) | | | | <0.001 |
| T1a | 54 (4.39) | 18 (2.05) | 36 (10.32) | |
| T1b | 345 (28.07) | 195 (22.16) | 150 (42.98) | |
| T1c | 376 (30.59) | 280 (31.82) | 96 (27.51) | |
| T2 | 454 (36.94) | 387 (43.98) | 67 (19.20) | |
| Radiotherapy | | | | <0.001 |
| No | 648 (52.73) | 363 (41.25) | 285 (81.66) | |
| Yes | 581 (47.27) | 517 (58.75) | 64 (18.34) | |
| Chemotherapy | | | | <0.001 |
| No | 446 (36.29) | 281 (31.93) | 165 (47.28) | |
| Yes | 783 (63.71) | 599 (68.07) | 184 (52.72) | |

SCLC, small cell lung cancer.

larger size of the tumor. In terms of therapy, the no-surgery group was more likely to have radiotherapy or chemotherapy. Given the unbalanced distribution of these factors between surgery and non-surgery groups, there is a need to reduce the interference from these factors to better determine the significance of surgery for prognosis in the older patients.

Univariate Cox analysis showed that the OS of patients was associated with age, T stage, surgery, radiotherapy, and chemotherapy (Figure S1A). Further multivariate Cox analysis showed that aged 65–70 years, right laterality, surgery, radiotherapy, and chemotherapy were the positive prognostic factors for OS (Figure S1C). Analogously, the variables of age, T stage, surgery, radiotherapy, and chemotherapy were related to the CSS of patients through univariate Cox analysis (Figure S1B). Age, laterality, surgery, radiotherapy, and chemotherapy were the independent predictive factors for CSS (Figure S1D). The Kaplan–Meier curves showed that the OS and CSS of patients aged ≥65 years with T1-2N0M0 SCLC who underwent surgery were significantly better than those who did not undergo surgery (both $P < 0.001$; Figure S2).

## Survival analysis and multivariate Cox analysis after propensity score matching

After 1:2 PSM of seven clinical characteristics, a total of 683 patients were included in the analyses, which contain 417 patients in the non-surgery group and 266 patients in the surgery group. The distribution of these baseline characteristics was balanced between the two propensity-matched groups (both $P > 0.05$; Table 2).

After PSM, the univariate Cox analysis showed that aged 65–70 years, surgery, radiotherapy, and chemotherapy were related to the OS of patients (Figure 2A), whereas aged 65–70 and 71–80 years, surgery, radiotherapy, and chemotherapy were associated with the CSS of patients (Figure 2B). Through further multivariate Cox analysis, the result showed that aged 65–70 and 71–80 years, tumors located on the left side, surgery, radiotherapy, and chemotherapy were the positive predictive factors for OS (Figure 2C); aged 65–70 and 71–80 years, tumors located on the right side, surgery, and radiotherapy were the independent prognostic factors for CSS (Figure 2D). The

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TABLE 2 The clinicopathologic characteristics of older patients with T1-2N0M0 SCLC after propensity score matching.

| Characteristics | Total (N,%) | No Surgery | Surgery | P-value |
|---|---|---|---|---|
| All | 683 | 417 | 266 | |
| Age (year) | | | | 0.175 |
| >80 | 92 (13.47) | 59 (14.15) | 33 (12.41) | |
| 65–70 | 233 (34.11) | 131 (31.41) | 102 (38.35) | |
| 71–80 | 358 (52.42) | 227 (54.44) | 131 (49.25) | |
| Sex | | | | 0.399 |
| Female | 383 (56.08) | 228 (54.68) | 155 (58.27) | |
| Male | 300 (43.92) | 189 (45.32) | 111 (41.73) | |
| Race | | | | 0.982 |
| Black | 31 (4.54) | 19 (4.56) | 12 (4.51) | |
| Other | 22 (3.22) | 13 (3.12) | 9 (3.38) | |
| White | 630 (92.24) | 385 (92.33) | 245 (92.11) | |
| Laterality | | | | 0.653 |
| Left | 291 (42.61) | 181 (43.41) | 110 (41.35) | |
| Right | 392 (57.39) | 236 (56.59) | 156 (58.65) | |
| T stage (eighth edition) | | | | 0.366 |
| T1a | 26 (3.81) | 15 (3.60) | 11 (4.14) | |
| T1b | 240 (35.14) | 139 (33.33) | 101 (37.97) | |
| T1c | 220 (32.21) | 133 (31.89) | 87 (32.71) | |
| T2 | 197 (28.84) | 130 (31.18) | 67 (25.19) | |
| Radiotherapy | | | | 0.054 |
| No | 497 (72.77) | 292 (70.02) | 205 (77.07) | |
| Yes | 186 (27.23) | 125 (29.98) | 61 (22.93) | |
| Chemotherapy | | | | 0.461 |
| No | 300 (43.92) | 178 (42.69) | 122 (45.86) | |
| Yes | 383 (56.08) | 239 (57.31) | 144 (54.14) | |

SCLC, small cell lung cancer.

Kaplan–Meier survival analysis after PSM showed that the OS of patients aged ≥65 years with T1-2N0M0 SCLC who underwent surgery were significantly better than those who did not undergo surgery ($P < 0.001$; Figure 3A). The median OS time of the surgery group was 35 months, which was significantly longer than the median OS time of the non-surgery group (13 months). After that, the median CSS time of the surgery group was also longer than that in the non-surgery group, with the median CSS time being 59 months in the surgery group and 14 months in the non-surgery group ($P < 0.001$; Figure 3B).

## Subgroup analysis of OS and CSS in subgroups of clinical characteristics

To better minimize the interference of other factors except for surgery on the prognosis and better determine the protective role of surgery on prognosis after PSM, we performed the subgroup analyses of all clinical characteristics. The OS subgroup analysis showed that the treatment of surgery was a protective factor for OS for almost clinical characteristics, except for the other subgroup of the race (Figure 4A). The CSS subgroup analysis showed that the surgical treatment was a protective factor for CSS for almost clinical characteristics, except for the other subgroup of race and the T1a subgroup of T stage (Figure 4B). The abovementioned clinical subgroups presented statistically insignificant differences in the OS or CSS between the surgery and no-surgery groups because the number of these subgroups was limited.

## Kaplan–Meier survival analysis of the OS and CSS for surgical treatment between different subgroups

To further determine the protective effect of surgical procedure on OS and CSS, we performed the subgroup analysis in different age, tumor size, and treatment groups after PSM. The result showed that the surgery group had a better prognosis than the non-surgery group regardless of OS or CSS, but the difference between the two surgery strategies' OS and CSS was not statistically significant in all age subgroups

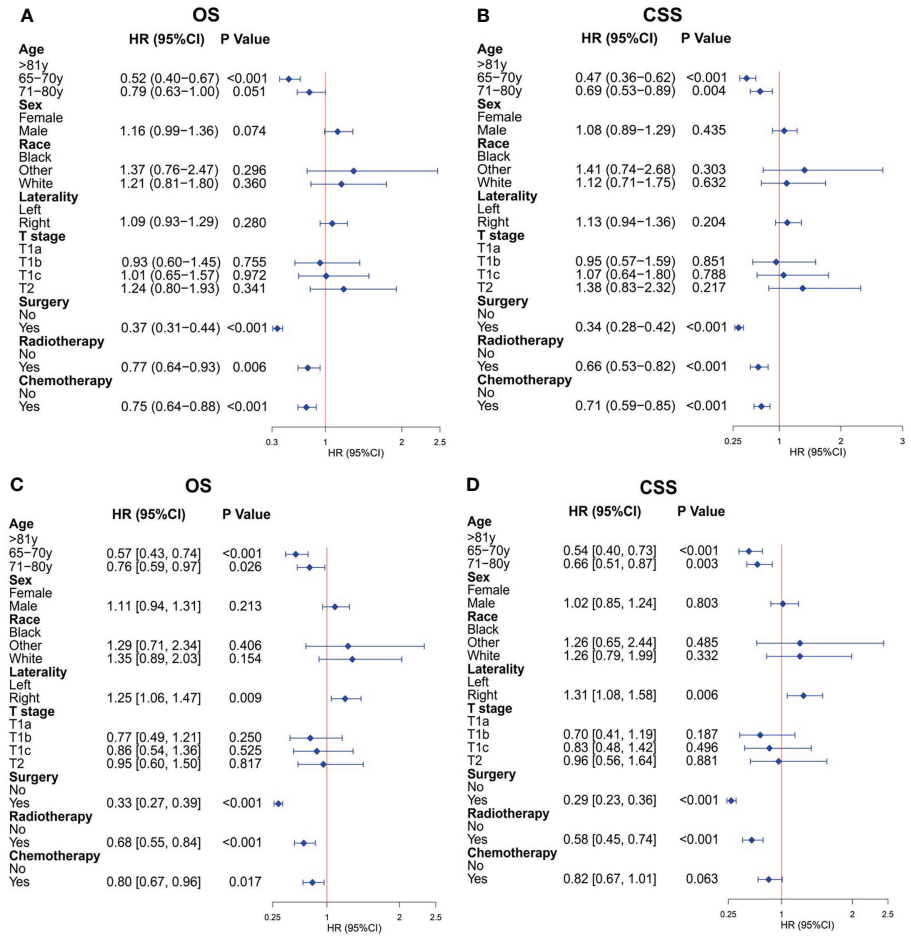


FIGURE 2
Cox regression analysis for overall survival (OS) and cancer-specific survival (CSS) of older patients with T1-2N0M0 small cell lung cancer (SCLC) after propensity score matching. **(A)** Univariate Cox analysis for OS. **(B)** Univariate Cox analysis for CSS. **(C)** Multivariate Cox analysis for OS. **(D)** Multivariate Cox analysis for CSS.

Figures 5A, B, D–F, except the OS in patients aged 71–80 years (Figure 5C, P = 0.024). However, all trends in survival benefits favored lobectomy over sublobectomy. In terms of the tumor size, we assembled the T1a and T1b as the group of T1a + T1b because the number of T1a group was limited with only 26 patients after PSM. The prognosis of two surgery strategies was better than that of no-surgery group in all T subgroups. The sublobectomy group had a worse prognosis than the lobectomy group in T1a + T1b stage subgroup regardless of OS or CSS (Figures 6A, D), whereas there was no significant difference in OS and CSS between the two different surgery strategies in the T2 subgroup (Figures 6C, F). In T1c subgroup analyses, the lobectomy group had a better prognosis than sublobectomy in OS, not in CSS, but the trend in survival benefit also favored lobectomy (Figures 6B, E). In terms of therapy, the surgery group all achieved better OS and CSS than the non-surgery group in patients who had chemotherapy alone (Figures 7A, E), radiotherapy alone (Figures 7B, F), and no chemotherapy or radiotherapy (Figures 7D, H), but the difference of prognosis in OS and CSS was insignificant between sublobectomy and no-surgery groups in patients who received chemotherapy plus radiotherapy (Figures 7C, G). For patients in the chemotherapy group, the lobectomy group could improve the prognosis in OS rather than CSS compared with the sublobectomy group, but the difference in OS and CSS of patients who received radiotherapy and chemotherapy plus radiotherapy was significant. However, this result was not well persuasive for the limited samples in the radiotherapy group. In no-chemotherapy or radiotherapy subgroup analyses, the OS and CSS of sublobectomy and lobectomy were comparable, but the outcomes were better than that of patients who did not undergo surgery (Figures 7D, H).

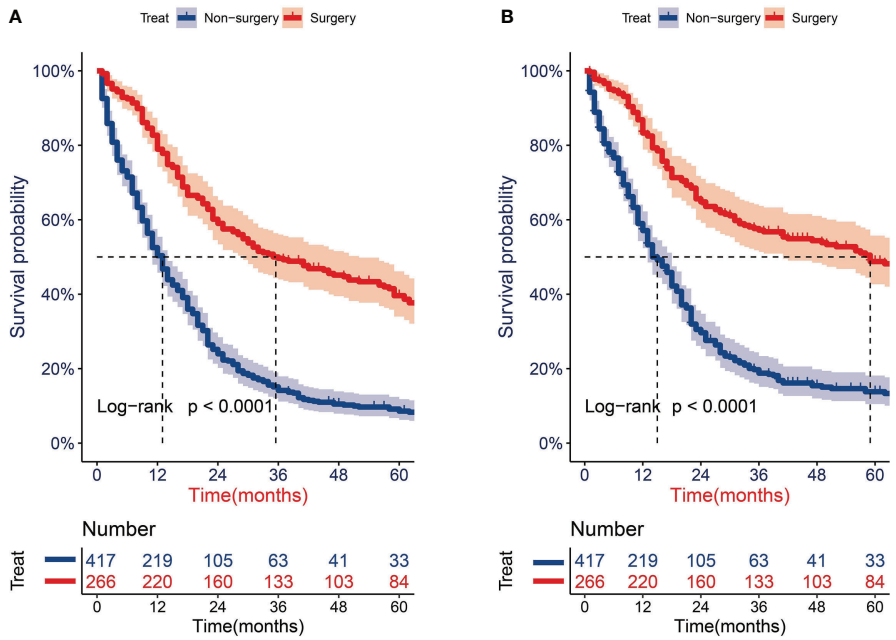


FIGURE 3
Survival analysis for overall survival (OS) and cancer-specific survival (CSS) of older patients with T1-2N0M0 small cell lung cancer (SCLC) after propensity score matching. **(A)** KM curves of OS. **(B)** KM curves of CSS.

## Prognostic factors of patients in the surgery group

To further explore the prognostic factors of older patients with T1-2N0M0 SCLC who underwent surgery, we performed the multivariate Cox analysis of the clinical characteristics of patients in the surgery group. The result presented that the characteristics of age (65–70 years), sex (female), and race (black) were the statistically positive influence factors for OS of patients (Figure 8A). After that, we find that just the factor of age (65–70 years) has a positive effect on the CSS of patients (Figure 8B).

# Discussion

The SEER database is currently the largest database of tumor clinical information in the world, which can help reduce the cancer burden among the US population. Many significant problems in clinical practice have been published using the SEER database in recent years (19, 20). However, the SEER database covers a long period and contains multiple different editions of the AJCC tumor staging system and other indicators, so it was so challenging to compare the results of the delivered research using the SEER database (21). Because of this reason, we converted the TNM staging of each patient into those of the eighth edition to guarantee that the study population information conformed to the current treatment guidelines. Notably, our study could provide credible and practical medical evidence for clinical decision-making of treatment in older patients with T1-2N0M0 SCLC through this approach.

The average age of patients diagnosed with SCLC increased, and the proportion of patients with SCLC older than 70 years had increased from 23% in 1975 to 44% in 2010 (22, 23). After that, the frequency of detecting early-stage lung cancer will be likely to increase as CT screening for lung cancer becomes more commonplace in recent years (24). Currently, the NCCN guidelines recommend surgery for selected cases of clinical stage T1-2N0M0 SCLC (25). However, considering the potential multiple comorbidities, increased treatment-related complications, decreased functional status, relatively high mortality in the older adult (26–29), and SCLC that is characterized by rapid growth and early metastasis, surgery is rarely performed in older patients even if their SCLC is at an early stage, and it was controversial whether the survival benefits of surgical treatment are significant for older patients. In our study, we also observed that the rate of surgery decreased with age increasing (34.6%, 27.4%, and 18.6% for the age subgroups 65–70, 71–80, and >80 years, respectively), and the surgical treatment could provide better prognosis than without surgery. Although there were several studies exploring the benefits of surgery in patients with early-stage SCLC, they did not stratify specifically by age and surgical procedure, and some of them had confounder interference (15, 30–32). Previous study also showed

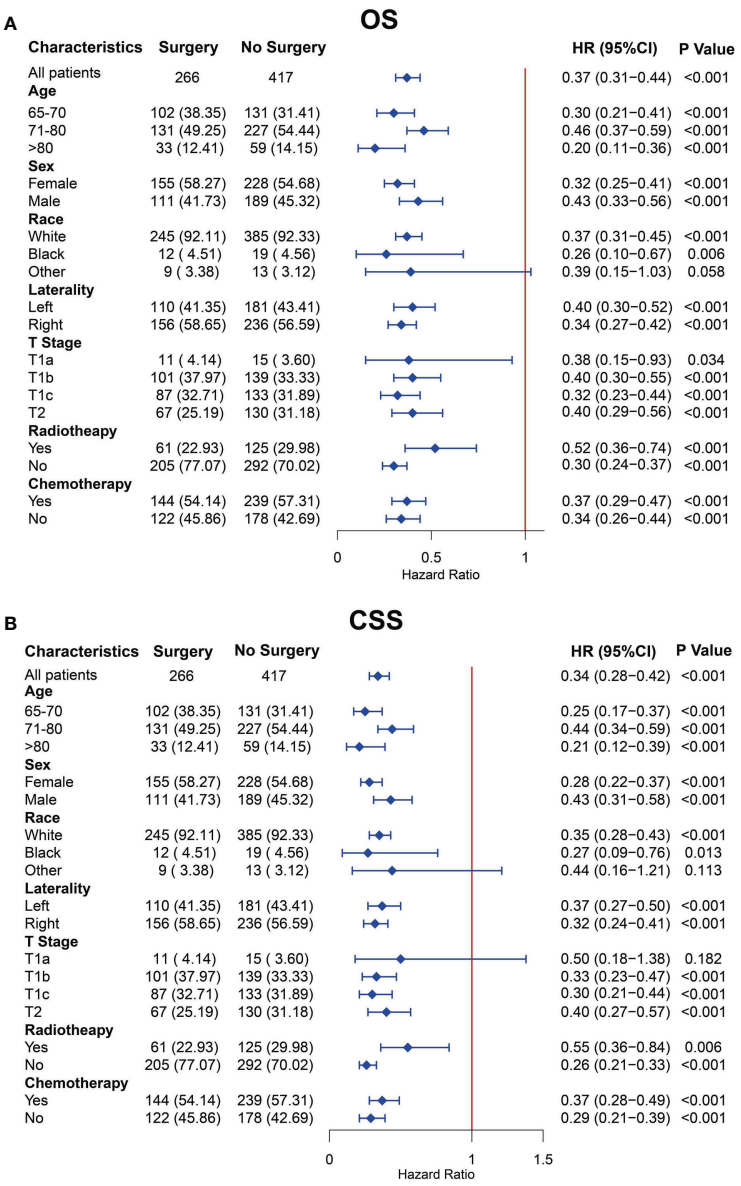


FIGURE 4
Subgroup analysis for overall survival (OS) **(A)** and cancer-specific survival (CSS) **(B)** of older patients with T1-2N0M0 small cell lung cancer (SCLC) after propensity score matching.

that age was the independent prognostic factor for patients with SCLC who received surgical treatment (33), which was consistent with the result of our research (Figure 8).

Before PSM, our results demonstrated that surgical treatment was the most significant protective factor of all clinical factors for OS and CSS, although a severe imbalance in the distribution of clinicopathological features between the surgery and non-surgery groups existed in our study. Whereas, the biases in data distribution in terms of baseline characteristics would interfere with the comparison between groups and the accuracy of the Cox regression model (34, 35). To determine the benefits of surgery in older patients and reduce confounding factor interference between the surgery group and the non-surgery group, we performed the 1:2 PSM to balance the distribution of a total of seven clinical characteristics so that the OS and CSS could be compared between the two groups at similar baselines and with a convincing result. After 1:2 PSM, with a total of 683 older patients with T1-2N0M0 SCLC, our results showed that the surgery remains the most important independent prognostic factor for older patients with T1-2N0M0 SCLC, and patients who underwent surgery achieved significantly better OS and CSS than those who did not undergo

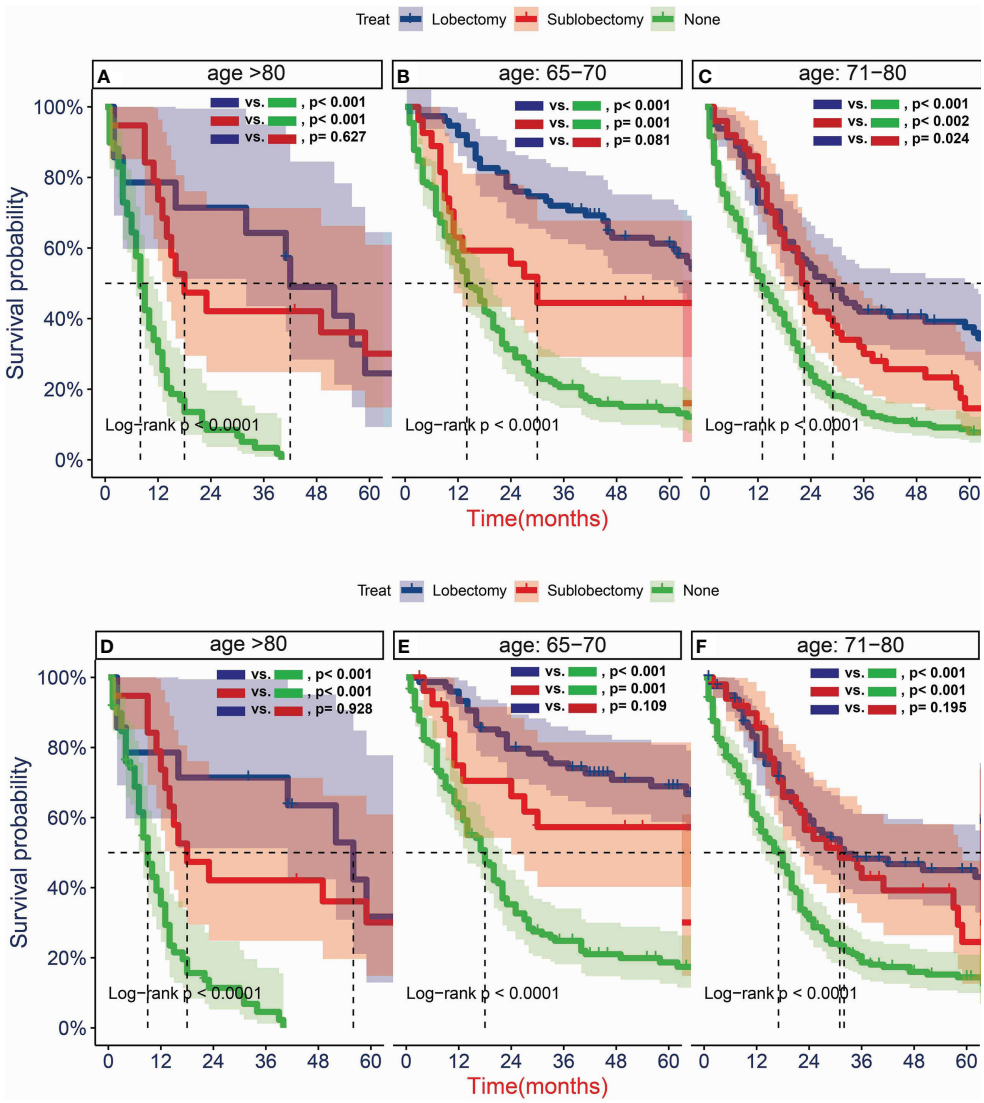


FIGURE 5
Kaplan–Meier estimates of overall survival (OS) **(A–C)** and cancer-specific survival (CSS) **(D–F)** for T1-2N0M0 patients with small cell lung cancer (SCLC) aged 65–70 years old **(B, E)**, 71–80 years old **(C, F)**, and >80 years old **(A, D)**stratified by surgery strategy after propensity score matching.

surgery ($P < 0.001$). Subgroup analysis also showed that surgical intervention was a protective factor for OS and CSS for almost clinical characteristics in older patients with SCLC. These results showed that a more aggressive treatment strategy may be beneficial in older patients with T1-2N0M0 SCLC, leading to a better survival of OS and CSS for these patients. Moreover, our study also noted that the factor of age 65–70 years was a protective factor of prognosis in older patients with SCLC undergoing surgery regardless of OS or CSS. Sex and race were independent predictors of OS in surgical patients and were not statistically significant in CSS, which is similar to the results of previous studies (15, 30, 32). Our study implied that these factors should be evaluated in detail before surgery and that intensive follow-up should be carried out for this special subset of patients although they have received surgical treatment.

The standard treatment for patients with limited-stage SCLC is chemotherapy and radiotherapy. However, considering the several physiological changes of organ function in older patients that could alter drug pharmacokinetics and have an impact on cytotoxic chemotherapy tolerability and toxicity, the treatment regimens may be different among different age (36). Ludbrook et al. analyzed retrospectively 174 patients with limited-stage SCLC and divided into three age groups: <65,

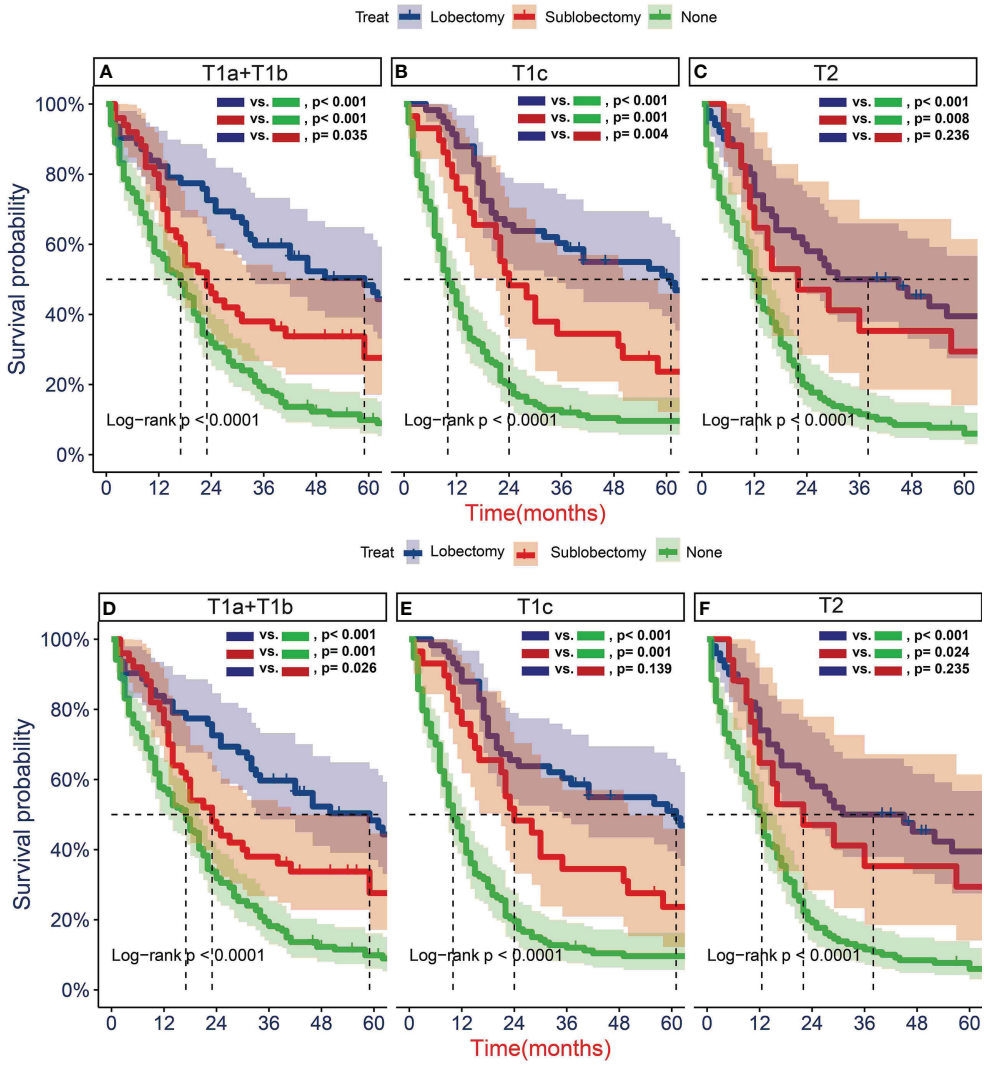


FIGURE 6
Kaplan–Meier estimates of overall survival (OS) **(A–C)** and cancer-specific survival (CSS) **(D–F)** for T1-2N0M0 patients with small cell lung cancer (SCLC) with stage T1a + T1b **(A, D)**, T1c **(B, E)**, T2 **(C, F)** stratified by surgery strategy after propensity score matching.

65–74, and ≥ 75 years. They found that increasing age was significantly associated with fewer diagnostic scans, less intensive chemotherapy regimens, fewer cycles, and lower total doses (37). In addition, there are some studies suggesting that the dose and frequency of radiotherapy were either less intensive in the elderly or comparable between younger and elderly patients (37, 38). After that, the local relapse occurs in up to 80% of limited-stage patients managed with chemotherapy alone, although SCLC was significantly sensitive to chemotherapy (39). Some data revealed that up to 16% of limited-stage SCLC died from a relapse confined to the thorax (40). Previous studies asserted that the treatment of operation could prevent local recurrence and improve survival in patients with SCLC (41, 42). A retrospective study published by Jin et al. also suggested that patients with T1-2N0 SCLC may benefit from surgery as local therapy, whereas patients with T3N0 or T1-2N1 SCLC may consider radiotherapy as local therapy (43). The American College of Chest Physicians and the American Society of Clinical Oncology also recommends surgery for patients with stage I SCLC, followed by adjuvant chemotherapy including platinum agent and etoposide (44, 45). In our study, we found that there was survival benefit for older patients who received surgery combined with chemotherapy or/plus radiotherapy compared with chemotherapy or/plus radiotherapy alone and that lobectomy may be the best choice, which was consistent with previous results (13, 42, 46). From the above results, it suggested that surgical treatment combined with adjuvant therapy may further improve the local control to prolong

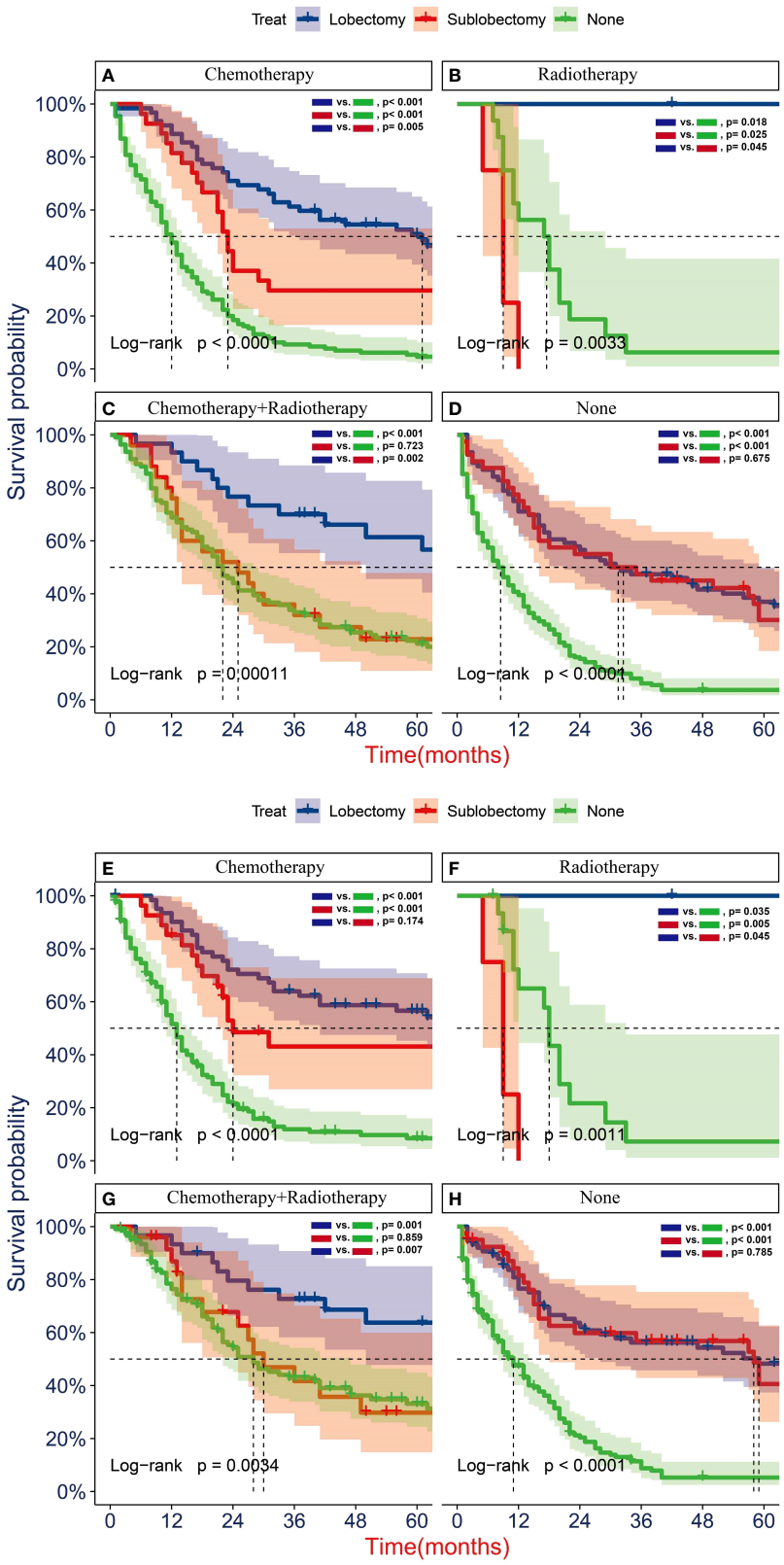


FIGURE 7
Kaplan–Meier analyses of overall survival (OS) **(A–D)** and cancer-specific survival (CSS) **(E–H)** for T1-2N0M0 patients with small cell lung cancer (SCLC) with chemotherapy **(A, E)**, radiotherapy **(B, F)**, radiotherapy plus chemotherapy **(C, G)**, and no radiotherapy or chemotherapy **(D, H)** stratified by surgery strategy after propensity score matching.

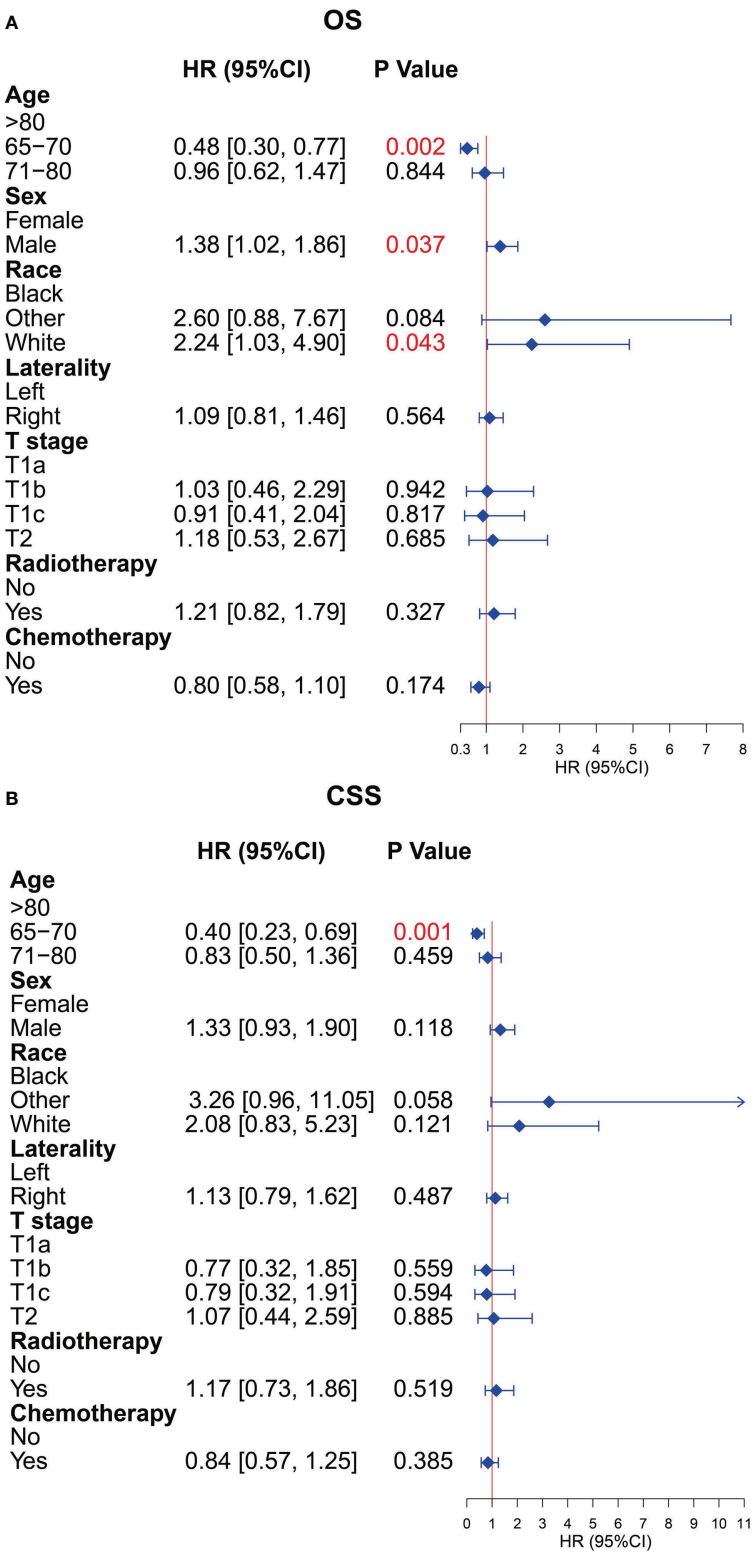


FIGURE 8
Multivariate Cox analysis for overall survival (OS) and cancer-specific survival (CSS) of the older patients with T1-2N0M0 small cell lung cancer (SCLC) who underwent surgery. **(A)** Multivariate Cox analysis for OS. **(B)** Multivariate Cox analysis for CSS.

survival and supported the role of surgery in multimodality therapy for older patients with T1-2N0M0 SCLC.

According to the NCCN guidelines, surgery is recommended for patients with T1-2N0M0 SCLC, and it points out that lobectomy is superior to sublobectomy (25). However, many patients with early-stage SCLC also undergo sublobectomy for various reasons (47). To the best of our knowledge, few studies have discussed whether sublobectomy can achieve the same survival outcomes comparable to lobectomy in older patients (≥ 65 years) with T1-2N0M0 SCLC and which type of surgery combined with adjuvant therapy is most effective for OS and CSS currently. In our study, we find that the trend in OS and CSS benefits favored lobectomy over sublobectomy, and all achieved better survival than patients without surgery, although there was generally no statistical difference between the two surgical procedures in almost age subgroups. These results presented that sublobectomy could be considered in older patients with SCLC when patients cannot tolerate lobectomy due to various reasons like multiple comorbidities or poor pulmonary function. In terms of tumor size, our study found that lobectomy was the priority choice compared with sublobectomy for tumors with tumor size less than 5 cm, because lobectomy could have longer OS and CSS than sublobectomy. The above results were similar to those of previous studies (30, 48). Meanwhile, we also found that surgery combined with chemotherapy plus/or radiotherapy could achieve better survival than chemotherapy plus/or radiotherapy alone, which is consistent with the result of the previous report (13). Moreover, patients who underwent lobectomy continued to have better survival in our study. Moreover, in the non-treatment subgroup, we find that the sublobectomy seems to achieve the same therapeutic effect as lobectomy regardless of OS and CSS and that the surgery group had a better prognosis than the patients without any therapy, which represented that the patients who just received the treatment of surgery could also achieve survival benefit as older patients do. The occurrence of this phenomenon was possibly associated with the poorer performance status or relatively short life expectancy of this population. For the patients with chemotherapy plus radiotherapy, these patients may have the high risk of metastasis and recurrence due to the larger tumor size or special location of the tumor. We find that the OS and CSS of patients with sublobectomy were comparable with that of patients in the no-surgery group, which achieved worse prognosis than patients with lobectomy. Thus, this special subset of older patients still could benefit from aggressive surgical treatment regardless of OS or CSS, and lobectomy should be the prior choice in older patients.

The current study had some limitations. First, as a retrospective study, the population selection may be biased inevitably and could not control for confounding factors as strictly as prospective studies. Although we have performed the PSM to reduce the potential bias, there might be a potential unknown bias that the PSM failed to rectify. After that, it is not clear how patients were selected for different treatment in the SEER database. Second, the SEER database lacked routinely available data including performance status, lung function, smoking status, and comorbidities. In particular, comorbidities could greatly influence treatment strategies and prognosis assessment and might be the reason why patients who undergo surgery have better survival than those who did not. Third, the information on the status of surgical margin, disease-free survival, chemotherapy regimen and cycles, radiotherapy dose and location, and further treatment after recurrence was not available. Moreover, we are uncertain whether these factors had an impact on our study, for which we should draw the conclusions carefully. In addition, the data in our study were extracted from the American population, and the results need to be verified using the data from Chinese population. Overall, further multicenter prospective studies with relatively complete information of clinicopathological variables, performance status, and treatments in detail should be performed to validate our conclusions and provide more reliable clinical guidance.

In conclusion, our study found that the long-term survival of older patients with T1-2N0M0 SCLC who received surgical treatment was significantly better than that of patients who did not undergo surgery after balancing all clinical characteristics and that lobectomy could provide a better prognosis than sublobectomy. For patients unsuitable for lobectomy, this special subset of patients also could benefit from sublobectomy. Age, sex, and race were independent prognostic factors of survival outcomes in older patients undergoing surgery. Therefore, the surgery should be performed for older patients with T1-2N0M0 SCLC after careful consideration and assessment combined with relevant clinical factors, but further exploration in larger prospective clinical trials is also needed to validate our conclusions.

## Data availability statement

The raw data supporting the conclusions of this article will be made available by the authors, without undue reservation.

## Author contributions

All authors participated in manuscript writing and approved the final version of the manuscript. JN, TG, YM, SZ conceived and designed the analysis. Collection and assembly of data were performed by JN, TG, SZ, YH, SR. Analysis and interpretation of the data were supported by JN, TG, SZ. YM, RL conducted a critical review of the manuscript, contributing important intellectual content.

## Conflict of interest

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

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## Supplementary material

The Supplementary Material for this article can be found online at: https://www.frontiersin.org/articles/10.3389/fonc.2022.958187/full#supplementary-material

## References

1. Bray F, Ferlay J, Soerjomataram I, Siegel RL, Torre LA, Jemal A. Global cancer statistics 2018: GLOBOCAN estimates of incidence and mortality worldwide for 36 cancers in 185 countries. *CA Cancer J Clin* (2018) 68(6):394–424. doi: 10.3322/caac.21492

2. Gazdar AF, Bunn PA, Minna JD. Small-cell lung cancer: What we know, what we need to know and the path forward. *Nat Rev Cancer* (2017) 17(12):725–37. doi: 10.1038/nrc.2017.87

3. Jackman DM, Johnson BE. Small-cell lung cancer. *Lancet* (2005) 366 (9494):1385–96. doi: 10.1016/S0140-6736(05)67569-1

4. Yu JB, Decker RH, Detterbeck FC, Wilson LD. Surveillance epidemiology and end results evaluation of the role of surgery for stage I small cell lung cancer. *J Thorac Oncol* (2010) 5(2):215–9. doi: 10.1097/JTO.0b013e3181cd3208

5. Schreiber D, Rineer J, Weedon J, Vongtama D, Wortham A, Kim A, et al. Survival outcomes with the use of surgery in limited-stage small cell lung cancer: Should its role be re-evaluated? *Cancer* (2010) 116(5):1350–7. doi: 10.1002/cncr.24853

6. Schild SE, Zhao L, Wampfler JA, Daniels TB, Sio T, Ross HJ, et al. Small-cell lung cancer in very elderly (>/= 80 years) patients. *Clin Lung Cancer* (2019) 20 (4):313–21. doi: 10.1016/j.cllc.2019.05.007

7. Gosney MA. Clinical assessment of elderly people with cancer. *Lancet Oncol* (2005) 6(10):790–7. doi: 10.1016/S1470-2045(05)70389-2

8. Fox W, Scadding JG. Medical research council comparative trial of surgery and radiotherapy for primary treatment of small-celled or oat-celled carcinoma of bronchus. *Ten-year follow-up. Lancet* (1973) 2(7820):63–5. doi: 10.1016/s0140-6736(73)93260-1

9. Pignon JP, Arriagada R, Ihde DC, Johnson DH, Perry MC, Souhami RL, et al. A meta-analysis of thoracic radiotherapy for small-cell lung cancer. *N Engl J Med* (1992) 327(23):1618–24. doi: 10.1056/NEJM199212033272302

10. Turrisi AT, Kim K, Blum R, Sause WT, Livingston RB, Komaki R, et al. Twice-daily compared with once-daily thoracic radiotherapy in limited small-cell lung cancer treated concurrently with cisplatin and etoposide. *N Engl J Med* (1999) 340(4):265–71. doi: 10.1056/NEJM199901283400403

11. Ganti AKP, Loo BW, Bassetti M, Blakely C, Chiang A, D'Amico TA, et al. Small cell lung cancer, version 2.2022, NCCN clinical practice guidelines in oncology. *J Natl Compr Canc Netw* (2021) 19(12):1441–64. doi: 10.6004/jnccn.2021.0058

12. Lim E, Belcher E, Yap YK, Nicholson AG, Goldstraw P. The role of surgery in the treatment of limited disease small cell lung cancer: time to reevaluate. *J Thorac Oncol* (2008) 3(11):1267–71. doi: 10.1097/JTO.0b013e318189a860

13. Yang CJ, Chan DY, Shah SA, Yerokun BA, Wang XF, D'Amico TA, et al. Long-term survival after surgery compared with concurrent chemoradiation for node-negative small cell lung cancer. *Ann Surg* (2018) 268(6):1105–12. doi: 10.1097/SLA.0000000000002287

14. Liu T, Chen Z, Dang J, Li G. The role of surgery in stage I to III small cell lung cancer: A systematic review and meta-analysis. *PloS One* (2018) 13(12): e0210001. doi: 10.1371/journal.pone.0210001

15. Li Y, Hu S, Xie J, Zhang X, Zong Y, Xu B, et al. Effects of surgery on survival of elderly patients with stage I small-cell lung cancer: analysis of the SEER database. *J Cancer Res Clin Oncol* (2019) 145(9):2397–404. doi: 10.1007/s00432-019-02976-2

16. Halvorsen TO, Sundstrom S, Flotten O, Brustugun OT, Brunsvig P, Aasebo U, et al. Comorbidity and outcomes of concurrent chemo- and radiotherapy in limited disease small cell lung cancer. *Acta Oncol* (2016) 55(11):1349–54. doi: 10.1080/0284186X.2016.1201216

17. Dunn C, Wilson A, Sitas F. Older cancer patients in cancer clinical trials are underrepresented. systematic literature review of almost 5000 meta- and pooled analyses of phase III randomized trials of survival from breast, prostate and lung cancer. *Cancer Epidemiol* (2017) 51:113–7. doi: 10.1016/j.canep.2017.11.002

18. Zhou B, Li Q, Qin L, Li Z, Jin K, Dai J, et al. Octogenarians may benefit from stage-specific small cell lung cancer treatment. *Transl Lung Cancer Res* (2021) 10 (10):3973–82. doi: 10.21037/tlcr-21-839

19. Yang L, Zhou Y, Wang G, Liu D, Chen B, Pu D, et al. Clinical features and prognostic factors of combined small cell lung cancer: development and validation of a nomogram based on the SEER database. *Transl Lung Cancer Res* (2021) 10 (11):4250–65. doi: 10.21037/tlcr-21-804

20. Zhu Z, Song Z, Jiao W, Mei W, Xu C, Huang Q, et al. A large real-world cohort study of examined lymph node standards for adequate nodal staging in early non-small cell lung cancer. *Transl Lung Cancer Res* (2021) 10(2):815–25. doi: 10.21037/tlcr-20-1024

21. Shao N, Xie C, Shi Y, Ye R, Long J, Shi H, et al. Comparison of the 7th and 8th edition of American joint committee on cancer (AJCC) staging systems for breast cancer patients: A surveillance, epidemiology and end results (SEER) analysis. *Cancer Manag Res* (2019) 11:1433–42. doi: 10.2147/CMAR.S185212

22. Abdel-Rahman O. Changing epidemiology of elderly small cell lung cancer patients over the last 40 years; a SEER database analysis. *Clin Respir J* (2018) 12 (3):1093–9. doi: 10.1111/crj.12632

23. Lally BE, Geiger AM, Urbanic JJ, Butler JM, Wentworth S, Perry MC, et al. Trends in the outcomes for patients with limited stage small cell lung cancer: An analysis of the surveillance, epidemiology, and end results database. *Lung Cancer* (2009) 64(2):226–31. doi: 10.1016/j.lungcan.2008.08.010

24. National Lung Screening Trial Research T, Aberle DR, Adams AM, Berg CD, Black WC, Clapp JD, et al. Reduced lung-cancer mortality with low-dose computed tomographic screening. *N Engl J Med* (2011) 365(5):395–409. doi: 10.1056/NEJMoa1102873

25. Kalemkerian GP, Loo BW, Akerley W, Attia A, Bassetti M, Boumber Y, et al. NCCN guidelines insights: Small cell lung cancer, version 2.2018. *J Natl Compr Canc Netw* (2018) 16(10):1171–82. doi: 10.6004/jnccn.2018.0079

26. Asmis TR, Ding K, Seymour L, Shepherd FA, Leighl NB, Winton TL, et al. Age and comorbidity as independent prognostic factors in the treatment of non small-cell lung cancer: a review of national cancer institute of Canada clinical trials group trials. *J Clin Oncol* (2008) 26(1):54–9. doi: 10.1200/JCO.2007.12.8322

27. Pal SK, Hurria A. Impact of age, sex, and comorbidity on cancer therapy and disease progression. *J Clin Oncol* (2010) 28(26):4086–93. doi: 10.1200/JCO.2009.27.0579

28. Okami J, Higashiyama M, Asamura H, Goya T, Koshiishi Y, Sohara Y, et al. Pulmonary resection in patients aged 80 years or over with clinical stage I non-small cell lung cancer: prognostic factors for overall survival and risk factors for postoperative complications. *J Thorac Oncol* (2009) 4(10):1247–53. doi: 10.1097/JTO.0b013e3181ae285d

29. Rostad H, Strand TE, Naalsund A, Talleraas O, Norstein J. Lung cancer surgery: the first 60 days. a population-based study. *Eur J Cardiothorac Surg* (2006) 29(5):824–8. doi: 10.1016/j.ejcts.2006.01.055

30. Liu Y, Shan L, Shen J, Liu L, Wang J, He J, et al. Choice of surgical procedure - lobectomy, segmentectomy, or wedge resection - for patients with stage T1-2N0M0 small cell lung cancer: A population-based study. *Thorac Cancer* (2019) 10 (4):593–600. doi: 10.1111/1759-7714.12943

31. Combs SE, Hancock JG, Boffa DJ, Decker RH, Detterbeck FC, Kim AW. Bolstering the case for lobectomy in stages I, II, and IIIA small-cell lung cancer using the national cancer data base. *J Thorac Oncol* (2015) 10(2):316–23. doi: 10.1097/JTO.0000000000000402

32. Weksler B, Nason KS, Shende M, Landreneau RJ, Pennathur A. Surgical resection should be considered for stage I and II small cell carcinoma of the lung. *Ann Thorac Surg* (2012) 94(3):889–93. doi: 10.1016/j.athoracsur.2012.01.015

33. Zeng Q, Li J, Tan F, Sun N, Mao Y, Gao Y, et al. Development and validation of a nomogram prognostic model for resected limited-stage small cell lung cancer patients. *Ann Surg Oncol* (2021) 28(9):4893–904. doi: 10.1245/s10434-020-09552-w

34. Yaya S, Gunawardena N, Bishwajit G. Association between intimate partner violence and utilization of facility delivery services in Nigeria: A propensity score matching analysis. *BMC Public Health* (2019) 19(1):1131. doi: 10.1186/s12889-019-7470-1

35. Gautier S, Monakhov A, Gallyamov E, Tsirulnikova O, Zagaynov E, Dzhanbekov T, et al. Laparoscopic left lateral section procurement in living liver donors: A single center propensity score-matched study. *Clin Transplant* (2018) 32 (9):e13374. doi: 10.1111/ctr.13374

36. Baker SD, Grochow LB. Pharmacology of cancer chemotherapy in the older person. *Clin Geriatr Med* (1997) 13(1):169–83.

37. Ludbrook JJ, Truong PT, MacNeil MV, Lesperance M, Webber A, Joe H, et al. Do age and comorbidity impact treatment allocation and outcomes in limited stage small-cell lung cancer? A community-based population analysis. *Int J Radiat Oncol Biol Phys* (2003) 55(5):1321–30. doi: 10.1016/s0360-3016(02)04576-5

38. Jara C, Gomez-Aldaravi JL, Tirado R, Meseguer VA, Alonso C, Fernandez A. Small-cell lung cancer in the elderly–is age of patient a relevant factor? *Acta Oncol* (1999) 38(6):781–6. doi: 10.1080/028418699432941

39. Albain KS, Crowley JJ, LeBlanc M, Livingston RB. Determinants of improved outcome in small-cell lung cancer: An analysis of the 2,580-patient southwest oncology group data base. *J Clin Oncol* (1990) 8(9):1563–74. doi: 10.1200/JCO.1990.8.9.1563

40. Jereczek B, Jassem J, Karnicka-Mlodkowska H, Badzio A, Mos-Antkowiak R, Szczepek B, et al. Autopsy findings in small cell lung cancer. *Neoplasma* (1996) 43(2):133–7.

41. Shepherd FA, Ginsberg RJ, Evans WK, Feld R, Cooper JD, Ilves R, et al. Reduction in local recurrence and improved survival in surgically treated patients with small cell lung cancer. *J Thorac Cardiovasc Surg* (1983) 86(4):498–506.

42. Badzio A, Kurowski K, Karnicka-Mlodkowska H, Jassem J. A retrospective comparative study of surgery followed by chemotherapy vs. non-surgical management in limited-disease small cell lung cancer. *Eur J Cardiothorac Surg* (2004) 26(1):183–8. doi: 10.1016/j.ejcts.2004.04.012

43. Jin K, Zhang K, Zhou F, Dai J, Zhang P, Jiang G. Selection of candidates for surgery as local therapy among early-stage small cell lung cancer patients: a population-based analysis. *Cancer Commun (Lond)* (2018) 38(1):5. doi: 10.1186/s40880-018-0272-5

44. Jett JR, Schild SE, Kesler KA, Kalemkerian GP. Treatment of small cell lung cancer: Diagnosis and management of lung cancer, 3rd ed: American college of chest physicians evidence-based clinical practice guidelines. *Chest* (2013) 143(5 Suppl):e400S–e19S. doi: 10.1378/chest.12-2363

45. Rudin CM, Ismaila N, Hann CL, Malhotra N, Movsas B, Norris K, et al. Treatment of small-cell lung cancer: American society of clinical oncology endorsement of the American college of chest physicians guideline. *J Clin Oncol* (2015) 33(34):4106–11. doi: 10.1200/JCO.2015.63.7918

46. Gu W, Zhong X. Efficacy of surgery combined with chemoradiotherapy in treating limited-stage small cell lung cancer and prognosis analysis. *J BUON* (2021) 26(3):812–8.

47. Wakeam E, Varghese TK Jr, Leighl NB, Giuliani M, Finlayson SRG, Darling GE. Trends, practice patterns and underuse of surgery in the treatment of early stage small cell lung cancer. *Lung Cancer* (2017) 109:117–23. doi: 10.1016/j.lungcan.2017.05.004

48. Okada M, Koike T, Higashiyama M, Yamato Y, Kodama K, Tsubota N. Radical sublobar resection for small-sized non-small cell lung cancer: a multicenter study. *J Thorac Cardiovasc Surg* (2006) 132(4):769–75. doi: 10.1016/j.jtcvs.2006.02.063

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# Integrated analysis of Helicobacter pylori-related prognostic gene modification patterns in the tumour microenvironment of gastric cancer

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**Background:** *Helicobacter pylori* (HP) infection is one of the leading causes of gastric cancer (GC). However, the interaction between HP and the TME, and its carcinogenic mechanism remains unknown.

**Methods:** The HP-related prognostic genes were identified based on HP infection-related gene markers and HP infection sample datasets by risk method and NMF algorithm. Principal component analysis (PCA) algorithm was used to constructed the HPscore system. The "limma" R package was employed to determine differentially expressed genes. In addition, the R packages, such as "xCell" and "GSVA", was used to analyze the relationship between the HPscore and tumor microenvironment. Finally, quantitative real-time polymerase chain reaction (qRT-PCR) was conducted to verify the expression levels of 28 HP-related prognostic genes in tissues.

**Results:** We successfully identified 28 HP-related prognostic genes that accurately classified the GC population. There are significant differences in survival between different subgroups (high-, low-risk and cluster_1,2). Thereafter, the HPscore system was constructed to evaluate the signatures of the 28 HP-related prognostic genes. The overall survival rate in the high-HPscore group was poor and immunological surveillance was reduced, whereas the low-HPscore group had a survival advantage and was related to the inflammatory response. HPscore was also strongly correlated with the tumour stage, TME cell infiltration and stemness. The qRT-PCR results showed

Abbreviations

HP, Helicobacter pylori; GC, Gastric cancer; BMDC, bone marrow-derived dendritic cells; CSC, cancer stem cells; DEGs, Differentially expressed genes; EMT, Epithelial-Mesenchymal Transition; GEO, Gene Expression Omnibus; TCGA, The Cancer Genome Atlas; GSVA, Gene Set Variation Analysis; HR, Hazard ratio; TME, Tumor microenvironment; GSEA, Gene Set Enrichment Analysis; ssGSEA, single sample Gene Set Enrichment Analysis; LASSO, Least absolute shrinkage and selection operator; OS, Overall survival; PPI, Protein–protein interaction; AUC, area under curve; KEGG, Kyoto Encyclopedia of Genes and Genomes; PCA, Principal component analysis

that DOCK4 expression level of 28 HP-related prognostic genes was higher in gastric cancer tissues than in adjacent tissues.
**Conclusions:** HP signatures play a crucial role in the TME and tumourigenesis. HPscore evaluation of a single tumour sample can help identify the TME characteristics and the carcinogenic mechanism of GC patients infected with HP, based on which personalized treatment can be administered.

KEYWORDS

*Helicobacter pylori*, gastric cancer, tumour microenvironment, cell stemness, prognostic model

# Introduction

Gastric cancer (GC) is the fifth-largest type of malignant tumor globally, and its high mortality makes it the third leading cause of cancer-related death (1). It is closely associated with Helicobacter pylori (HP) infection. The World Health Organisation has listed HP as the first group of carcinogens causing gastric adenocarcinoma (2). Because HP is not an intracellular pathogen, continuous inflammation does not effectively eliminate HP but leads to epithelial cell damage. Further, the constant production of reactive oxygen species continues to cause DNA damage, which initiates the cascading reactions that lead to cancer development (3). A study showed that HP eradication therapy reduces the risk of GC in patients with first-degree relatives who have a family history of GC (4). Unfortunately, most patients are prone to drug resistance against HP and the infection cannot be eradicated. A recent observational study confirmed that HP infection can be completely eradicated in only 35% of patients who receive the follow-up treatment for this infection (2). In addition, the understanding of the carcinogenic mechanism of HP is still not comprehensive. Increasing evidence has suggested that the accumulation of bone marrow-derived dendritic cells (BMDCs) induced by HP is one of the origins of GC stem cells. Chronic HP infection leads to chronic inflammation and subsequent gastric epithelial mucosal damage, leading to the recruitment of BMDCs (5). BMDCs exhibit the phenotype and characteristics of cancer stem cells (CSCs) and obtain the ability to differentiate into gastric epithelial cells possibly through cell fusion (6, 7). This mechanism involves the secretion of various cytokines by infected epithelial cells, of which tumour necrosis factor-α (TNF- α) plays a significant role mainly through the NF-kB-dependent pathway (8). HP has been known to activate the typical NF-kB signal in gastric epithelial cells, and its mechanism depends on the type IV secretory system (T4SS) encoded by the CagA pathogenicity island of HP (9). Simultaneously, the inflammatory response caused by HP makes the tumour microenvironment (TME) more complex. With the transition from acute inflammation to chronic inflammation, the virulence factors released by HP prevent the differentiation of immune killer cells and promote the accumulation of immunosuppressive cells (9). In addition, HP activates tumour-associated fibroblasts by activating the IL-17 pathway to assist tumour cells in immune escape (10, 11). Further, the accumulation of a large number of fibroblasts makes it difficult for immune cells to enter the tumour core and provides the necessary conditions for angiogenesis. Therefore, identifying the characteristics of HP-mediated gastric epithelial cell infiltration can help in strengthening our understanding of the complex and changeable TME.

In this study, we identified the prognostic gene markers associated with HP infection in patients with GC. These genes showed a strong correlation with tumour immune-infiltrating cells, and to some extent, participated in the signal pathway of tumour stem cells and then affected tumour progression. We constructed an HPscore system by using HP-related prognostic genes to comprehensively evaluate the TME modification patterns in patients with GC. Elucidation of the overall mechanism of HP infection can help us understand its carcinogenic nature and develop effective treatment strategies.

# Materials and methods

## Collection and preprocessing of datasets

The flowchart (**Figure 1**) and mechanism diagram were plotted in the BioRender (https://app.biorender.com). First, we retrieved HP-related studies published in the past 3 years from the NCBI and Web of Science to verify the HP-infected related gene markers (the following unified abbreviated as HP). To investigate the relationship between HP infection and GC, we collected relevant datasets from the GEO and TCGA databases. In summary, 4 HP infection-related datasets (i.e. GSE6143, GSE5081, GSE27411, and GSE60662), 5 GC datasets (i.e., GSE66229, GSE29272, GSE84437, GSE15459, and TCGA-STAD) with OS data, three drug treatment datasets (i.e., PD-L1/IMvigor210). The ROC curve and the AUC value were used to evaluate the diagnostic efficacy of HP-related genes. The "survival" and "survminer" R packages were used to draw the survival curve of the GC datasets. To eliminate the batch effects of different datasets, we used the "combat" algorithm of the "SVA" R package to merge the datasets (i.e., GSE66229 and

GSE15459). The "FactoMineR" and "Factoextra" R packages were used to demonstrate the fit effect of the meta-dataset. The "Corrplot" R package was used to identify the potential HP regulatory genes in the meta-datasets. We used the "limma" R package to determine the differential genes between Hp-positive and Hp-negative patients in the GEO dataset (i.e. GSE6143 and GSE60662). The "upsetR" and "VennDiagram" R packages were employed to identify the overlapping genes. Then, the univariate Cox regression analysis was performed to identify HP-related prognostic genes, and the HR values of these genes were visualised using the "forestplot" R package.

## Evaluation of the clinical value of HP-related prognostic genes

Based on the HP-related prognostic genes, lasso regression and multivariate Cox regression were used to establish the prognostic risk model with the "survival" and "glmnet" R packages. Then, the samples were classified into high- and low-risk groups according to the median risk score. The "pheatmap", "survival", and "survminer" R packages were employed to demonstrate the difference in the prognosis between the high- and low-risk groups. The "scatterplot3d" R package was applied to investigate the distribution of patients with a different risk score.

## Nonnegative matrix factorisation

To evaluate the modification differences among the GC samples, we used the Nonnegative Matrix Factorisation (NMF) method to classify 482 GC patients from the meta-datasets based on the presence of HP-related prognostic genes. When the decreasing trend of the cophenetic correlation coefficient was most obvious, the k value was regarded as the best cluster number. The "NMF" R package was employed to plot the heatmap, basis components, and the connectivity matrix of NMF in different clusters.

## PPI network and functional pathway enrichment analysis

The protein–protein interaction network was constructed using the Search Tool for the Retrieval of Interacting Genes database (STRING, https://string-db.org/). Cytoscape software with the MCODE plugin was employed for the optical network and to identify the most significant module. The GO function annotation and the KEGG pathway enrichment analysis were performed using the "clusterProfiler" R package and DAVID (https://david-d.ncifcrf.gov/). The signal pathway gene sets were downloaded from MSigDB (https://www.gsea-msigdb.org/gsea/msigdb). Gene enrichment analysis was also performed using GSEA software (version 4.0).

## Generation of HP-related prognostic gene signature

To quantify the HP-related prognostic gene modification patterns in each sample, we defined the HPscore, a scoring system for evaluating individual GC patients. The principal component analysis (PCA) was performed to construct the HPscore. Similar to that described in previous studies (12, 13), we added PC1 and PC2 as the final gene signature scores. The HPscore was represented as

$$\text{HPscore} = \sum_{i}^{j} (PC1i + PC2i)$$

The samples were categorised as high- and low-HPscore groups, with the optimal cutoff value. In addition, the distribution of patients with the HPscores was visualised using the t-distributed random neighbour embedding (T-SNE) method ("Rtsne" R package).

## Estimation of TME and stemness feature

The "xCell" R package was used to calculate the microenvironment score for the meta-dataset. In addition, the ESTIMATE was used to calculate tumour purity and the immune infiltration levels (14). Thus, a comprehensive microenvironment score that reflected tumour purity and immune cell infiltration in the tumour samples was constructed. According to the markers of immune cells obtain from the Charoentong's research (15), the single-sample gene-set enrichment analysis (ssGSEA) algorithm was employed to quantify the relative abundance of each immune cell infiltration in the GC tumour microenvironment by using the "GSVA" R package, and each immune cell infiltration score was standardised for further analyses. We also used the biological pathways constructed by Mariathasan et al. (16) to evaluate the association between the HPscore and biological processes, including (1) immune checkpoint; (2) antigen processing machinery (APM); (3) epithelial–mesenchymal transition (EMT) markers such as the EMT1, EMT2, and EMT3; (4) angiogenesis signature; (5) pan fibroblast TGF-b response signature (Pan-FTBRS); and (6) CD8+ T-effector signature. All the gene sets used in the study are listed in the **Supplementary Table S6**.

## Tissue samples and quantitative real-time polymerase chain reaction

A total of 24 tumor tissue and 20 normal adjacent tissue were collected from patients with GC. Following are the

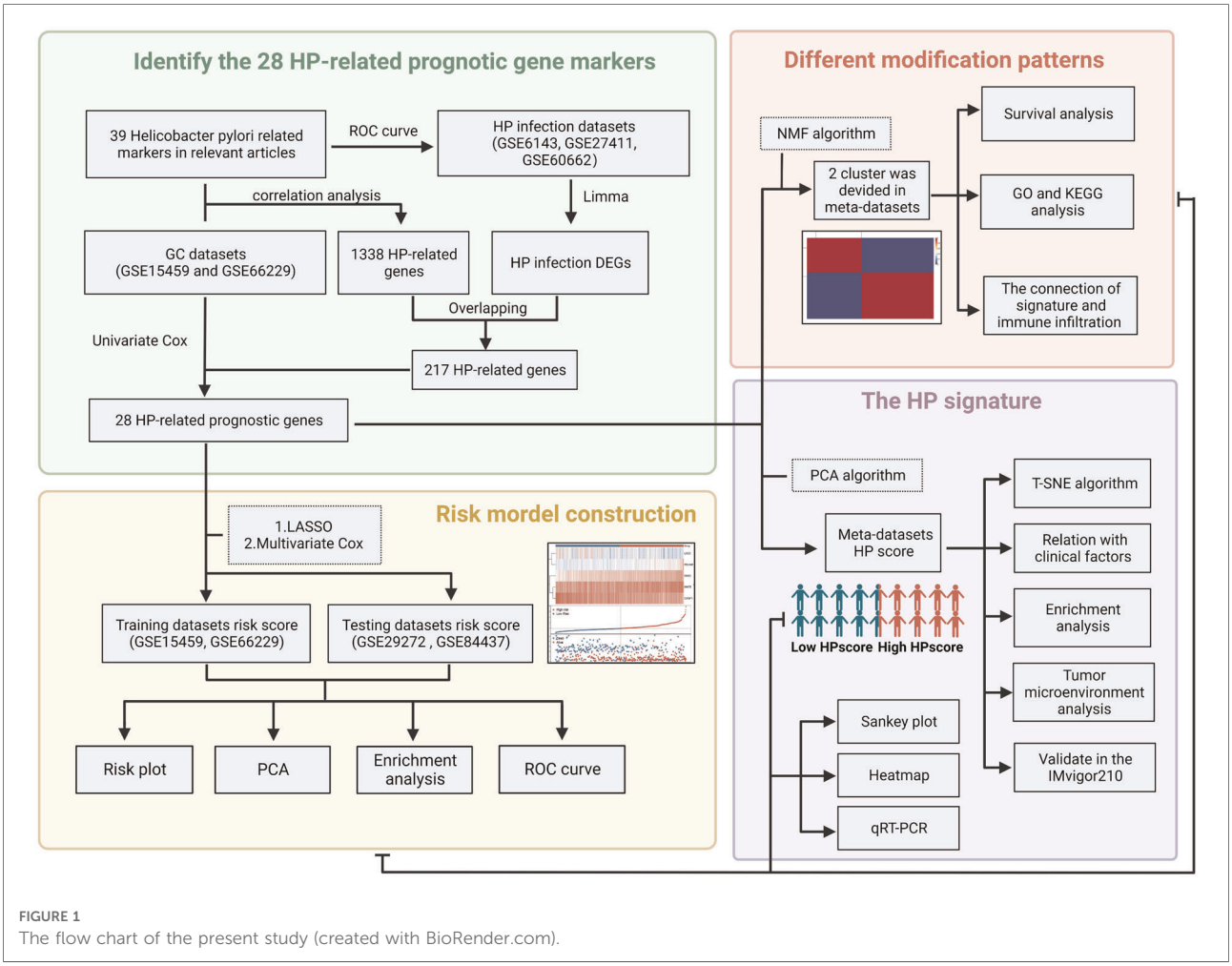


FIGURE 1
The flow chart of the present study (created with BioRender.com).

inclusion criteria for tissue specimens: (1) Diagnosis of GC from a pathological perspective; (2) Except for GC without other malignancies; (3) Surgical procedures are not preceded by radiotherapy or chemotherapy. The study was approved by the Ethics Committee and informed written consent was obtained from all patients. The specific experimental protocol for qRT-PCR referred to our previous research methods.

## Statistical analysis

All the data were processed using R 4.0.1 software. We obtained mutation data of the GC samples from the TCGA database. The "maftools" R package was employed to visualise mutation data. Independent prognostic factors were identified through the Cox analysis. The "limma" R package was employed to determine differentially expressed genes (DEGs) between the subgroups with fold change = 1, and the volcano map was used for visualisation. Survival curves were generated using the Kaplan–Meier method, and log-rank tests were performed to calculate the differences. The Sankey diagram was developed using the "networkD3" R package. The "ggplot2", "ggpubr", and "pheatmap" R packages were used to visualise the results. Pearson correlation coefficient among the data were calculated through the "Corrplot" R package and visualised using the "PerformanceAnalytics", "Hmisc", and "ggstatsplot" R packages. All statistical *P*-values were two-sided, and a *P* value of <0.05 was considered to be statistically significant. *P*-values: ns, $P > 0.05$; *$P < 0.05$; **$P < 0.01$; ***$P < 0.001$; ****$P < 0.0001$.

# Results

## The landscape of HP-related genes in GC

Through systematic literature screening, a total of 39 genes were considered to be gastric infection HP gene signature (**Supplementary Table S1**), termed as HP-related genes. ROC results from three different datasets (GSE6143, GSE27411, and GSE60662) suggested that the HP-related gene sets can effectively diagnose HP infection (**Supplementary Figure S1A**). **Figure 2A** shows the dynamic carcinogenic process induced by

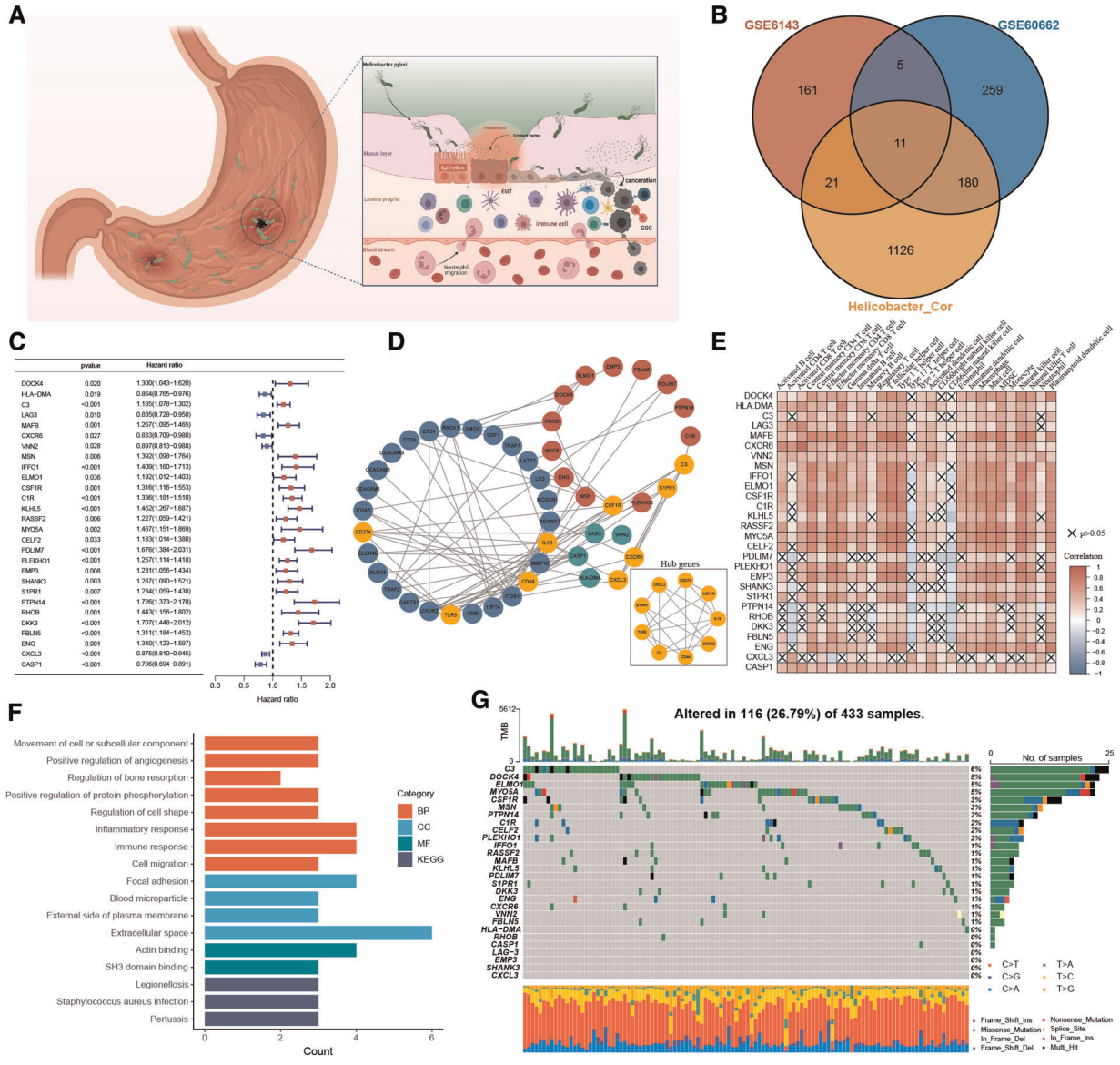


FIGURE 2
The landscape of HP-related prognostic genes in gastric cancer. (**A**) Pathogenesis of Helicobacter pylori. (**B**) Venny diagram displaying the overlap of differential gene expression profiles. (**C**) Blue and red represented the hazard ratio by the univariate cox regression model. (**D**) Protein-protein interaction (PPI) network (Blue nodes represent HP-related genes, Green and red are HP-related prognostic genes. Orange represents the hub genes identified by the mcode method). (**E**) Visualization of the correlation analysis results between HP-related prognostic genes and tumor-infiltrating immune cells in the meta-dataset. (**F**) GO and KEGG analysis results of 28 HP-related prognostic genes. (**G**) The mutation information of 28 HP-related prognostic genes was analyzed in the TCGA-STAD cohort.

HP infection in the stomach. To determine the best cluster dataset, we first performed the survival analysis to evaluate prognostic differences between the datasets (**Supplementary Figure S1B**). After excluding the datasets with poor data quality, GSE15459 and GSE66229 were integrated into training datasets (**Supplementary Figures S1C,D**), and TCGA-STAD and GSE84437 were used as testing datasets. Subsequently, the correlation analysis was performed to determine the correlation between the HP-related genes and meta-dataset, and a total of 1,338 genes were identified (**Supplementary Table S2**). The Venn plot was used to show the overlapping region between 1,338 genes and the differential genes in GSE6143 and GSE60662. A total of 217 genes were confirmed for follow-up analysis (**Figure 2B** and **Supplementary Figure S2A**). To further determine the relationship between HP infection and GC, the univariate Cox regression model showed that 28 genes were associated with GC prognosis (**Figure 2C**), and these genes were termed as HP-related prognostic genes. A protein

interaction network of 21 HP-related prognostic genes and the HP-related genes was constructed. The mcode plugin was used to identify potential hub genes (Figure 2D). The strong correlation among the HP-related prognostic genes is shown in Supplementary Figures S2B. In addition, Spearman correlation analysis showed a robust correlation between HP-related prognostic genes and tumour immune-infiltrating cells (Figure 2E). Type 17 helper T cells and CD56 dim natural killer cells were negatively correlated with poor prognostic genes but positively correlated with favourable prognostic genes. Subsequent KEGG and GO enrichment analyses also confirmed the strong correlation between these prognostic genes and the immune signal pathway. For example, the "inflammatory response" and "immune response" signalling pathways of GO were enriched. In addition, the enrichment of "positive regulation of angiogenesis" and "focal adhesion" and "cell migration" signalling pathways indicated the potential biological function of 28 prognostic genes in GC (Figure 2F). We studied the mutation incidence in TCGA-STAD patients to fully describe the characteristics of HP prognostic genes (Figure 2G). Of the 433 samples, 116 harboured a mutation in HP-related prognostic genes, with a frequency of 26.79%. *C3*, *DOCK4* and *ELMO1* had the highest mutation rate (6%), followed by *MYO5A* (5%). These results suggested that the HP-related prognostic genes are strongly associated with the immune microenvironment of GC and tumour progression.

## Risk stratification of patients with gastric cancer based on HP-related prognostic genes

We performed lasso regression of 28 genes based on meta-dataset, and the results suggested that *LAG3*, *MAFB*, *PDLIM7*, *DKK3*, and *CASP1* can be used to establish risk models (Figure 3A). Multivariate Cox analysis was then used to calculate the risk score of each sample in the meta-dataset. The forest plot showed the relationship between the five genes and cancer prognosis (Figure 3B). Then, GC patients were divided into high- and low-risk groups, with the median risk score as the threshold. Survival analysis showed that the survival time of the high-risk group was significantly lower than that of the low-risk group (Figure 3C, $P < 0.0001$). The heatmap result showed that *MAFB*, *PDLIM7*, and *DKK3* were highly expressed in the high-risk group but *LAG3* and *CASP1* were not expressed. With the increase in the risk score, the proportion of death in patients increased significantly (Figure 3D). Principal component analysis showed significant differences in the high- and low-risk cohorts (Figure 3E). The area under the curve (AUC) of the meta-datasets at 1, 2, and 3years were 0.68, 0.70, and 0.68 (Figure 3F). The datasets GSE29272 and GSE84437 were used to evaluate the actual value of the risk model of the high- and low-risk cohorts, respectively. These results suggested that our model can well stratify the risk of GC, and a significant difference was observed in the survival of GC patients between the high- and low-risk groups (Supplementary Figure S3A). As shown in Figure 3G, the high-risk group had a high levels of regulatory T cells, T follicular helper cells, type 1 T helper cells, mast cells, and plasmacytoid dendritic cell infiltration in the tumour rather than activated CD4 T cells, activated CD8 T cells, type 17 helper T cells, and CD56 light/dim natural killer cells. We used GSEA to analyse the enrichment level of the pathways. The hallmark and GO enrichment pathways, including Wnt, autophagy, TGF-B, EMT, Angiogenesis, Hypoxia, Notch, and Hedgehog signalling pathways were of considerable attention (Figure 3H) because the participation and imbalance of these pathways might be the reasons for the poor prognosis in the high-risk group. Significant differences were found in HP-related gene expression between high- and low-risk groups (Supplementary Figure S3B).

To summarise, the risk model based on 28 HP-related prognostic genes can be used as an essential index to evaluate the prognosis of GC. At the same time, multiple tumour-related signal pathways were enriched. Significant differences were found in the expression of HP-related genes and the distribution of infiltrating immune cells among the two risk groups.

## Different modification patterns of HP-related prognostic genes

The risk stratification of the population was successfully performed by building the risk model. We then classified the patients based on meta-dataset by using the NMF method, calculated the NMF symbiotic correlation coefficient, and selected $k = 2$ as the best grouping value (Supplementary Figures S4A,B). We successfully obtained two different modification patterns of HP-related prognostic genes in patients with GC, termed cluster_1 and cluster_2 (Figures 4A, B). Significant differences were observed in the survival between cluster_1 and cluster_2 (Figure 4C, $P < 0.0001$). A total of 338 DEGs were identified in the two HP-related prognostic gene modification groups (Figure 4D and Supplementary Table S3). The clusterProfiler R package was used to identify the function and signalling pathways of differential genes. The results showed that bacterial invasion- and inflammation-related pathways were enriched in cluster_1, indicating favourable prognosis (Figures 4E,F). The enrichment pathways of cluster_2 were mainly extracellular matrix- and membrane protein receptor-related signaling pathways (Figure 4G). The distribution of infiltrating immune cells in cluster_1 and cluster_2 was different (Figure 4H). Specifically, cluster_1 was rich in the infiltrated cells involved in inflammatory stress, such as activated CD4 T cells, type 17 helper T cells, activated dendritic cells, CD56 dim natural killer

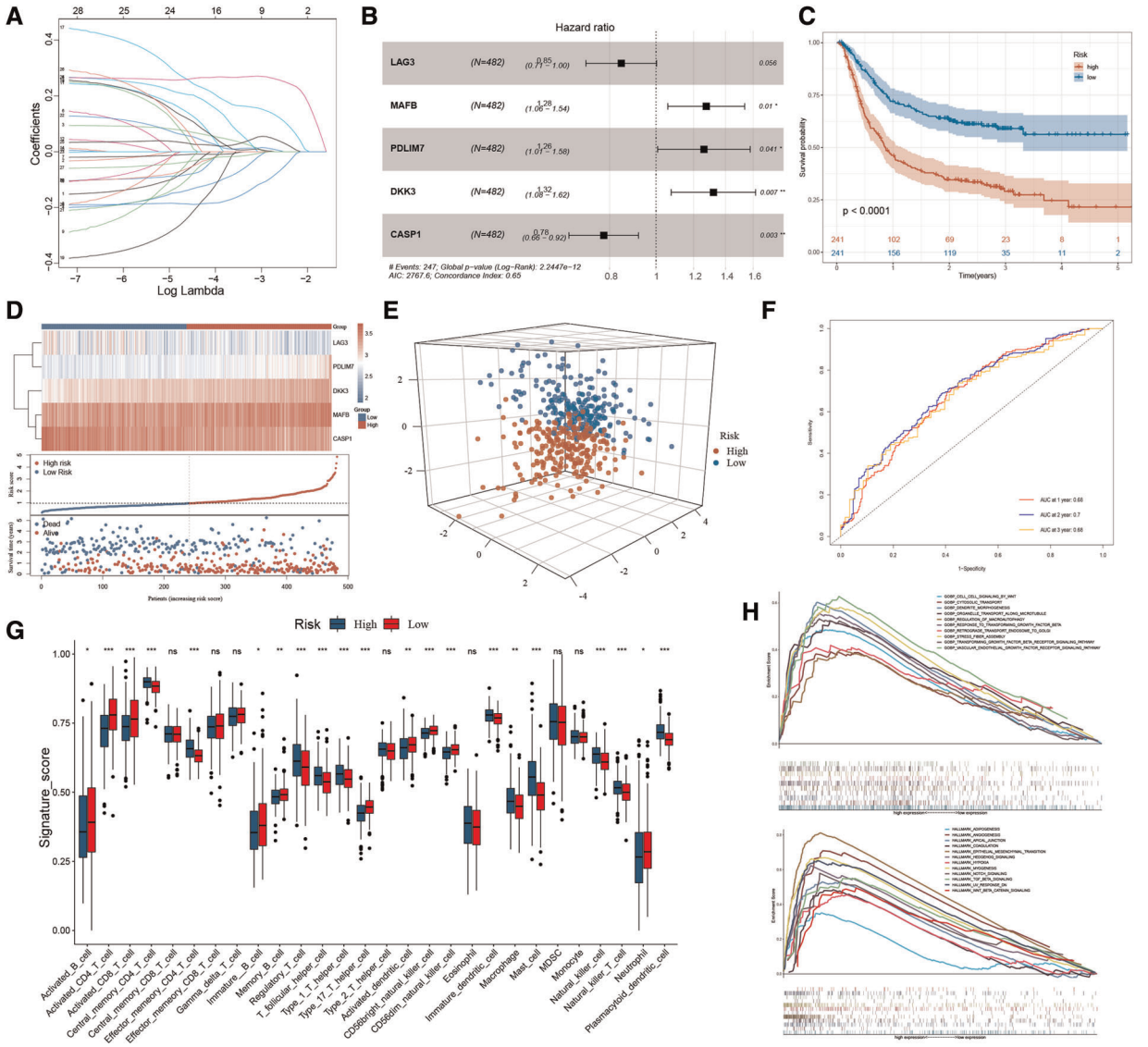


FIGURE 3
The workflow of risk-score model construction. (**A**) LASSO model coefficients. (**B**) *$P < 0.05$ in multivariable Cox proportional hazards model. (**C**) Kaplan-Meier survival curve of the OS in the high- and low-risk groups (meta-dataset). (**D**) From top to bottom are five prognostic signature RNAs expression heatmap, the risk score, and patients' survival status distribution between low- and high-risk groups. (**E**) Principal component analysis (PCA) shows the difference between the high-risk and low-risk groups based on the risk score. (**F**) ROC curves for 1-year, 2-year and 3-year overall survival, with AUC = 0.68 , 0.70 and 0.66 respectively. (**G**) The box plot results suggest that tumor-infiltrating immune cells were significantly differently distributed in the high-risk and low-risk groups. (**H**) GSEA results show the relevant signaling pathways involved in the high-risk group.

cells, and neutrophils. In contrast, cluster_2 showed infiltration of adaptive immune cells including regulatory T cells, T follicular helper cells, type 1 T helper cells, macrophages, mast cells, and plasmacytoid dendritic cells. Importantly, the epithelial cell signalling pathway in HP infection was enriched in cluster_1. Subsequently, we verified the expression of HP-related genes between cluster_1 and cluster_2 (**Supplementary Figure S4C**). These results directly confirm the reliability of the HP-related prognostic gene. A total of 28 HP-related prognostic genes can accurately classify the population of GC patients, and a significant difference was observed in tumour progression between cluster_1 and cluster_2.

## Generation of a HP-related prognostic gene signature

To further explore the biological differences in HP-related prognostic genes among individual GC samples, we constructed an HPscore system by employing the PCA

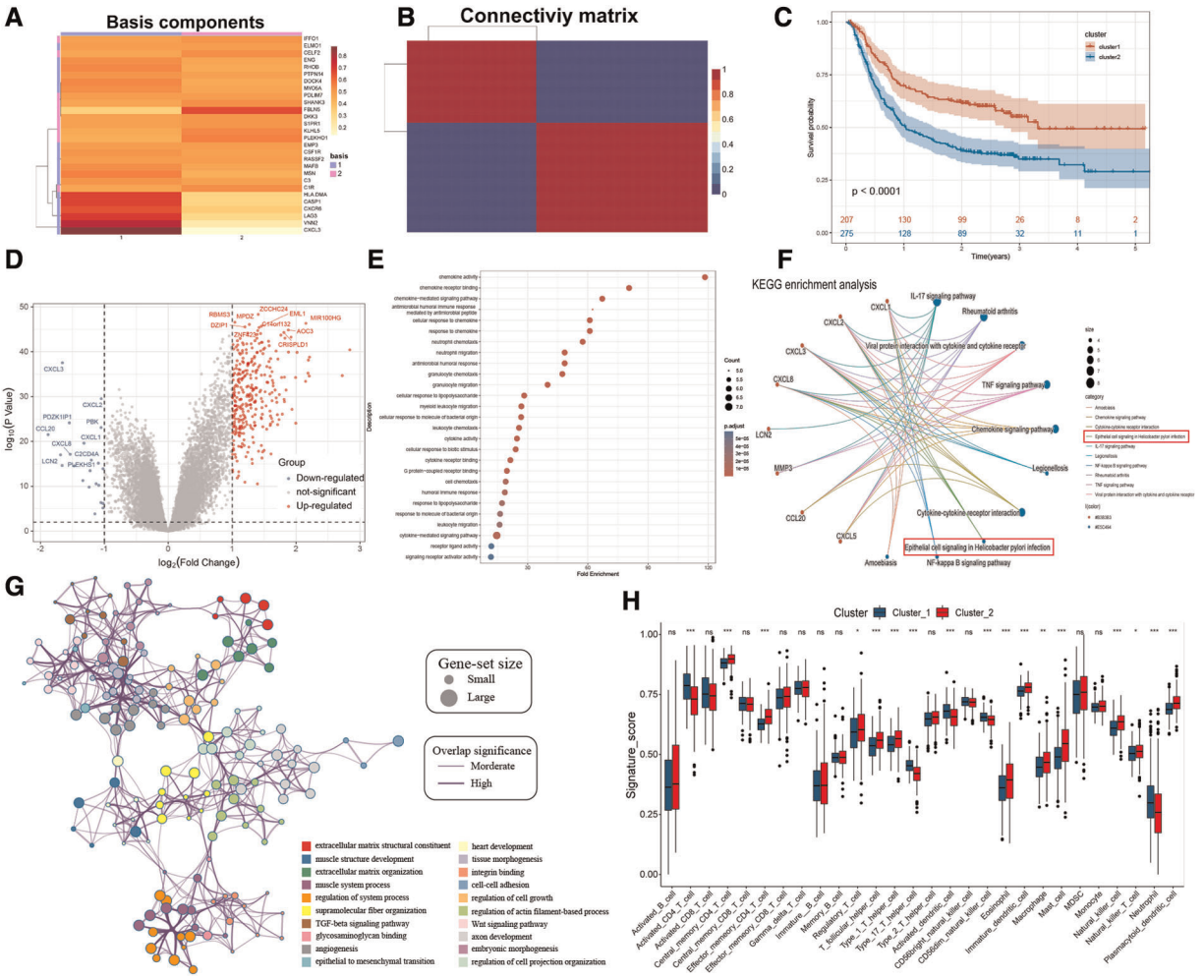


FIGURE 4
NMF for HP-related prognostic genes modification patterns, biological processes, and immune cell infiltration analysis. (A) NMF heatmap of basic components of HP-related prognostic genes expression in the two clusters. (B) Connectivity matrix for patients with gastric cancer in the meta-dataset by NMF. (C) Kaplan-Meier curves showing survival differences between two clusters. (D) Volcano plot shows the different genes between cluster 1 and cluster 2. (E,F) Bubble plot and gene-concept network shows up-regulated genes in cluster 1 involved signaling pathways. (G) Cytoscape and enrichment maps are used to visualize the function enrichment analysis results of up-regulated genes in cluster 2 involved signaling pathways. (H) Box plot results show differences in the distribution of infiltrating immune cells in different clusters.

method based on meta-datasets (Figure 5A and Supplementary Figure S5A). The T-SNE algorithm was used to visualise the sample HPscore (Figure 5B,C), and the results showed an apparent distance gradient among the GC samples with the increase in the HPscore. The meta-dataset was divided into two groups based on the optimal cutoff value: the high Hp_Score group ($n = 212$) and the low Hp_Score group ($n = 256$). Similar to the risk model, the high Hp_Score group demonstrated a shorter survival time than the low Hp_Score group (Figure 5D, $P < 0.00001$). To assess the stability and expansibility of the scoring system, the HPscores between the internal datasets GSE15459 and GSE66229 were compared, and no significant difference was observed between the two datasets (Supplementary Figure S5B). External datasets GSE29272 (GPL96) and GSE84437 (GPL6947) were used for the verification of the survival analysis, and the results are shown in Supplementary Figure S5C ($P = 0.004$) and S5D ($P = 0.009$). We then summarised the clinical information in meta-datasets to verify the relationship between the HPscore and clinical features (Supplementary Table S4). The results suggested that the HPscore increased with the increase in the TNM stage of cancer (Figure 5E); similar results were obtained through internal grouping (Figure 5F). We then analysed differences in the HPscores between the high- and low-risk groups and between cluster_1 and cluster_2 (Supplementary Figures S5E,F). The area under the curve (AUC) of the meta-datasets at 1, 2, and 3years were 0.64, 0.67, and 0.64 (Figure 5G), respectively. The alluvial diagram shows the flow of modified samples with

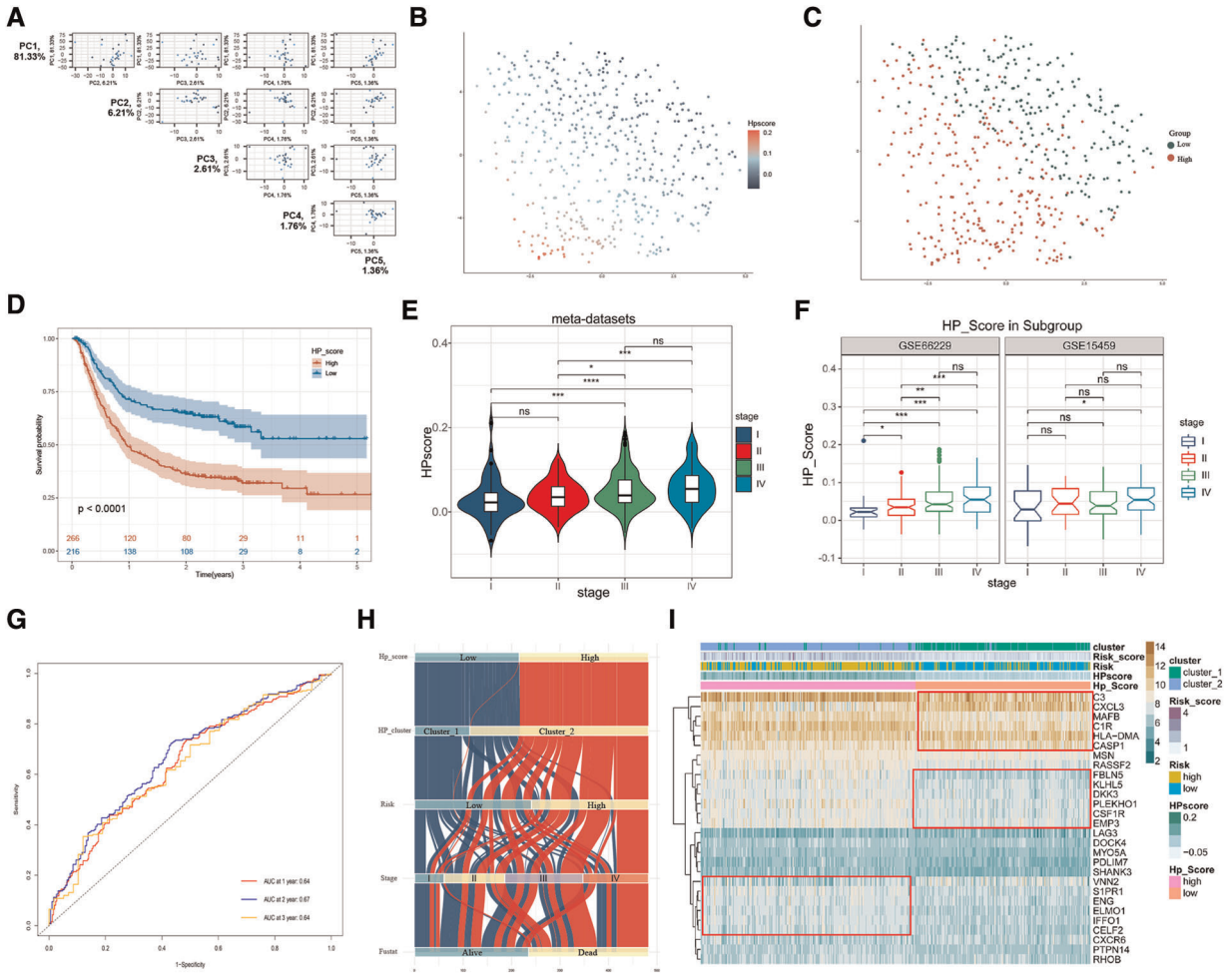


FIGURE 5
Construction of HPscore system and verification of system stability. (**A**) Pairs plot showing the results of principal component analysis (PCA) base on 28 HP-related prognostic genes. (**B**,**C**) The T-SNE of HPscore and Hp_Score groups for all samples. (**D**) Overall survival analysis verified HPscore system in meta-dataset. (**E**,**F**) The differences of HPscore in gastric cancer stages were observed in the meta-dataset and internal datasets, respectively. (**G**) ROC curves for 1-year, 2-year and 3-year overall survival, with AUC = 0.64, 0.67 and 0.64 respectively. (**H**) The Sankey map of samples in HP_score group, risk group, clusters group, stages, and survival outcome. (**I**) Heatmap of the expression levels of 28 HP-related prognostic genes in different groups.

different HPscores in a risk group, cluster group, stage cluster, and survival cluster (Figure 5H). We performed unsupervised cluster analysis based on 28 HP-related prognostic genes to determine the relationship between different GC subgroups (Figure 5I). The results showed a high degree of consistency between HPscore, Risk, and Cluster, and significant differences in the expression of three gene subgroups between the groups.

## Relationship between the HPscore and immune microenvironment

To confirm the relationship between HPscore and immune infiltration, we scored the samples by using the ssGSEA and xCell method. Activated CD4 T cells, type 17 T helper cells, TNF signalling, and IFNA signalling were mainly enriched in the low Hp_Score, whereas regulatory T cells and macrophages were significantly increased in the high Hp_Score (Figure 6A). The correlation analysis showed that the HPscore was significantly and positively correlated with the matrix cell score ($r = 0.74$, $P < 0.0001$) (Figure 6B) and microenvironment score ($r = 0.41$, $P < 0.001$) (Figure 6C). The comprehensive landscape of stromal cells in the high- and low-Hp score groups is shown in the heatmap (Figure 6D). The number of epithelial cells in the high Hp_Score group was found to be significantly lower than that in the low Hp_Score group, whereas the number of fibroblasts and endothelial cells increased significantly in the high HP group. To study the relationship between the HPscore and inflammation, we first verified the relationship between the

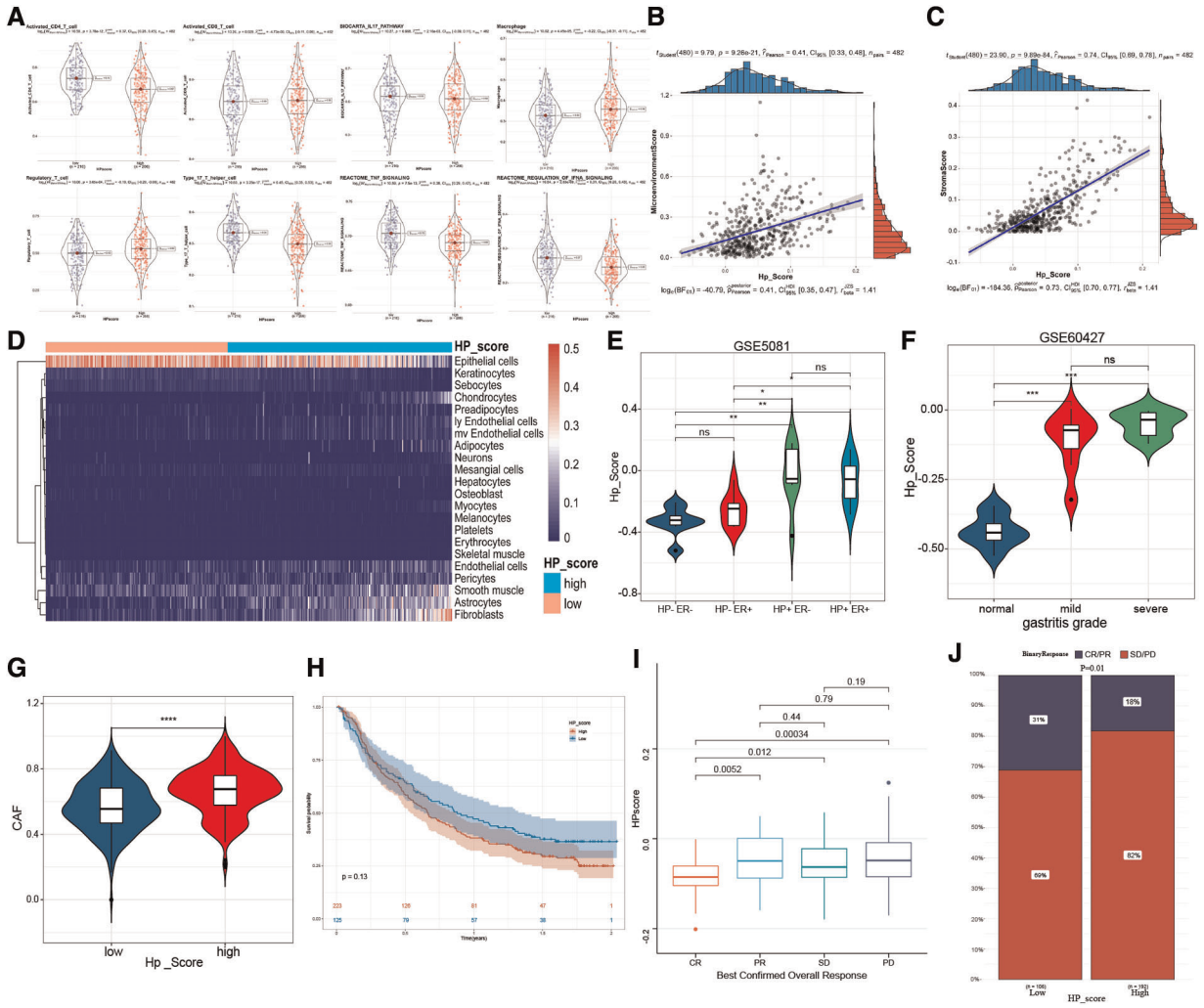


FIGURE 6
The immune-related characteristics of HPscore. (A) Significant differences of immune cell infiltration between high-HPscore and low-HPscore groups. (B,C) A scatter plot of the positive relationship between HPscore and Stromastore and Microenvironmentscore. (D) The heatmap shows the different distribution of stromal cells between both groups. (E,F) The significant differences of HPscore in patients with different levels of HP infection. (G) Relative distribution of CAF cells in HPscore high vs. low subgroups. (H) Kaplan-Meier curves for high and low HPscore patient groups receiving anti-PDL1 treatments. (I,J) The fraction of patients with clinical response to anti-PDL1 immunotherapy in low or high HPscore groups.

HPscore and Hp infection in the GSE5081 dataset. Regardless of inflammation, the Hp-positive group had a higher HPscore than the Hp-negative group (Figure 6E). In the dataset of inflammation induced by Hp infection (GSE60427), the HPscore better reflected the level of inflammation (Figure 6F, $P < 0.001$). To evaluate the relationship between the HPscore and cancer-related fibroblasts (CAF), we scored meta-dataset samples based on CAF cell characteristic genes. The results showed that the CAF enrichment score in the high Hp_Score group was significantly higher than that in the low Hp_Score group (Figure 6G, $P < 0.00001$).

Immunotherapy is a significant breakthrough in tumour therapy. We further explored the relationship between the HPscore and immunotherapy in the immunotherapy cohorts IMvigor210 (Supplementary Table S5). We found that the survival rate of patients with a high Hp_Score was lower than that of patients with a low Hp_Score, and the response to treatment was worse in the high Hp_Score group (Figures 6H–J).

## Analysis of the relationship between the HPscore and tumour stemness of GC

The study of the biological processes of tumor progression related to the HPscore showed that the HPscore

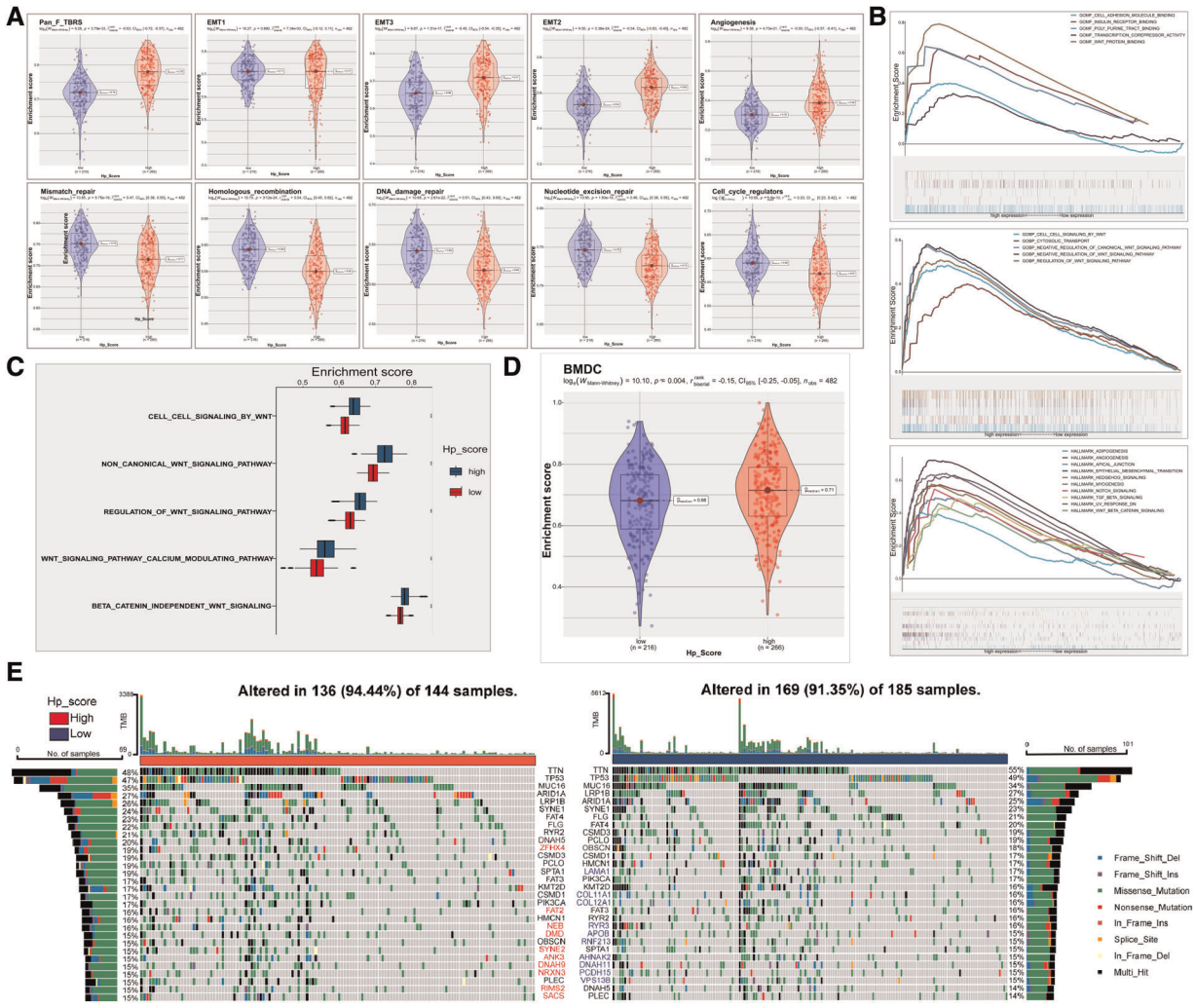


FIGURE 7
Clinical features and tumor stemness signature of HPscore. (A) Boxplot for the significant differences of the current immune-related signatures between two HP_score groups. (B) Prediction of HPscore-related pathways and molecular function by enrichment plots from GO analysis and hallmark enrichment pathway. (C) Differences enrichment scores of Wnt pathway between two HP_score groups. (D) Boxplot for the significant differences of BMDC enrichment scores between the two HP_score groups. (E) The landscape of tumor somatic mutation between the two HP_score groups.

was positively correlated with EMT2, EMT3, Pan_f_TBRS, and angiogenesis in GC but negatively correlated with DNA damage repair, mismatch repair, homologous recombination, nucleotide excision repair, and cell cycle regulators (Figure 7A). GSEA functional enrichment analysis suggested that EMT, Angiogenesis, cell adhesion, extracellular matrix junction, Wnt, TGF-b, Hedgehog, and Notch pathway were widely enriched in the high Hp_Score group (Figure 7B). Subsequently, we compared the performance of gene sets related to the Wnt pathway in the HPscore subgroup. The enrichment score of the high Hp_Score group was higher than that of the low Hp_Score group (Figure 7C). Similar results were obtained using BMDC enrichment scores (Figure 7D). Finally, the somatic mutation map in the TCGA cohort showed no significant difference in the mutation rates of *TP53*, *TTN*, and other top 30 genes between the high Hp_Score group and the low Hp_Score group (Figure 7E).

## Validation of HP-related prognostic genes

The TCGA-STAD dataset was used to verify the expression of 28 HP-related prognostic genes. The results showed that

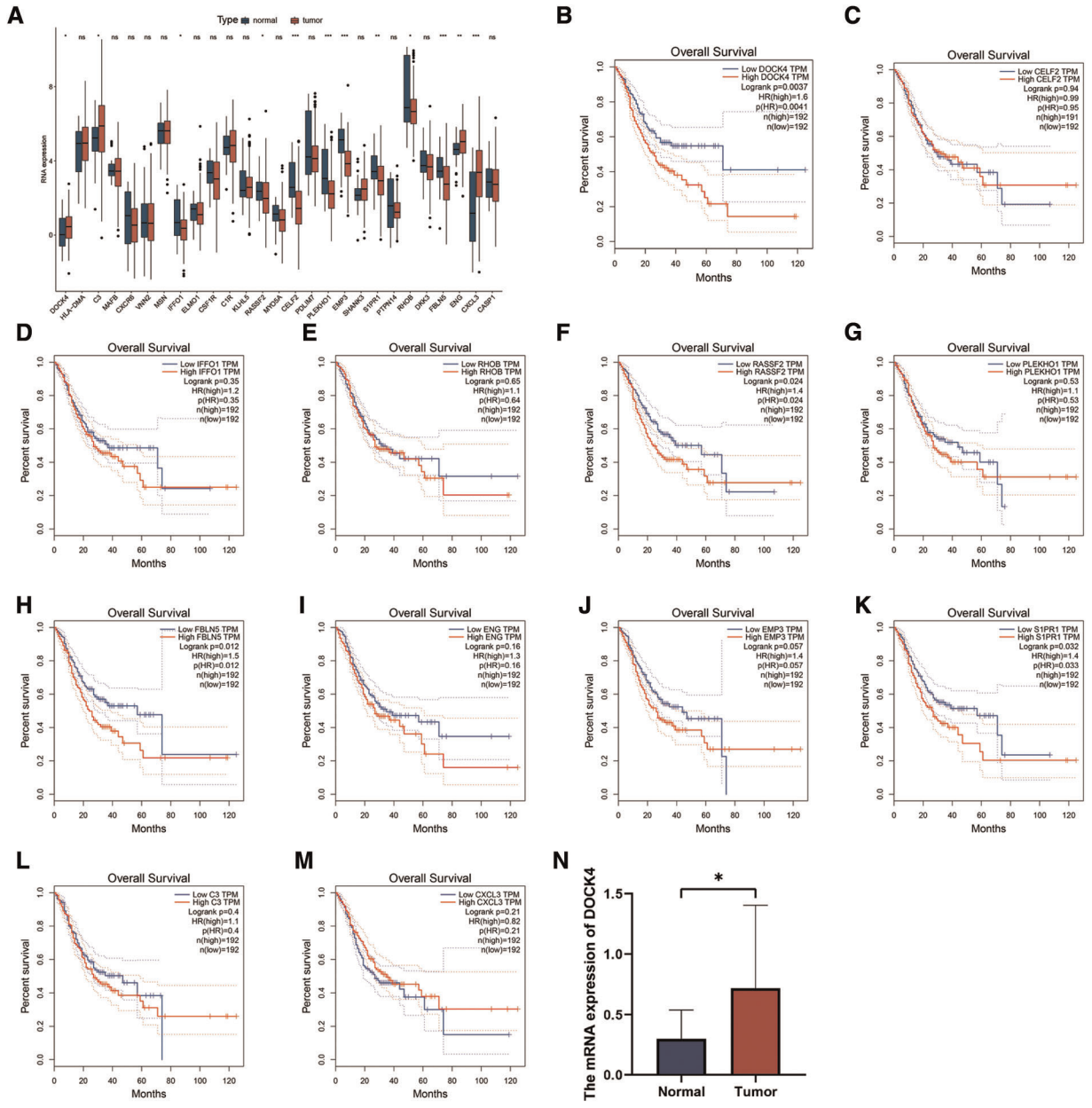


FIGURE 8
Expression and validation of 28 HP-related prognostic genes. (A) The expression patterns of 28 HP-related prognostic genes were analyzed in the TCGA-STAD cohort. (B–M) Analysis of overall survival with HP genes high and low expression groups in TCGA-STAD cohort. (N) qRT-PCR analysis of the expression of DOCK4 in 29 normal and 24 tumor tissue samples.

DOCK4, C3, ENG, and CXCL3 were highly expressed in patients with TCGA-STAD tumours, whereas IFFO1, RASSF2, CELF2, PLEKHO1, EMP3, S1PR1, RHOB, FBLN5, and CASP1were highly expressed in healthy individuals (Figure 8A). Survival analysis in TCGA-STAD cohort show that the patients with high expression of DOCK4, RASSF, FBLNF or S1PR1 had poorer overall survival (Figures 8B–M, $P < 0.05$). The results of qRT-PCR indicated that the mRNA level of DOCK4 was higher in carcinoma tissues than those in normal tissues (Figure 8N, $P = 0.0126$).

## Discussion

HP infection is the most common risk factor for GC. The virulence factors produced by HP affect the signal

transmission between cells, and chronic infection of gastric mucosa leads to changes in the local microenvironment. Most people infected with HP do not show symptoms related to bacterial virulence, host genetic polymorphism, and environmental factors (1). First, we identified the genetic markers associated with HP infection in patients with gastropathy by searching for HP-related literature. Because of the lack of sample data on large-scale carcinogenesis caused by HP infection, we used the correlation analysis to identify the gene modification phenotype of HP-related genes in GC. By performing systematic analysis, 28 genes were identified for follow-up research. By constructing a prognostic risk model and through NMF grouping, we identified two clinical tags of 28 genes: "prognostic indicators" and "HP infection associated." Based on the results, we considered 28 genes as HP-related prognostic genes in GC. Then, we created an HPscore system to quantify the modification characteristics of these 28 HP-related prognostic genes in the samples and determined the accuracy and stability of the HPscore system by using external datasets. After stringent verification, we concluded that the HPscore system can accurately reflect the status of HP infection and survival outcome in patients with GC. To confirm our inference, we successfully divided the patients with GC into two subgroups based on differences in the HPscore. Differences in the survival outcomes and HP infection status between subgroups were significant, which increased our confidence in continuing using HPscore to explore the detailed mechanisms of HP pathogenesis.

To the best of our knowledge, this study is the first to determine the relationship between HP infection and the 39 genetic markers identified through the ROC curve analysis. Our results showed that the areas under the ROC curve of the three datasets were >0.7, indicating that the 39 gene sets could accurately reflect the status of HP infection. To further explore the role of HP infection in GC, we performed correlation analysis to identify potential regulatory genes of HP in GC based on the meta-dataset. To improve the association between these genes and HP, we used HP+ and HP− differential gene datasets to screen the regulatory gene sets used in the previous step. Subsequently, the survival data were introduced into the analysis and by using univariate Cox analysis, we identified 28 genes for follow-up research. The results of the protein interaction network suggested that *C3*, *CSFR*, *S1PR1*, *CXCR6*, and *CXCL3* could be primarily involved in HP pathogenesis. HP cytotoxin-associated gene A (CagA) has been reported to relieve the inhibitory effect of *TGF- β* on *CXCL3* and aggravate the inflammatory response (17). Studies have reported that *S1PR1* is associated with the differentiation of memory T cells (18–20) and affects the prognosis of GC by promoting chemotherapy resistance (21, 22). As expected, the 28 genes were strongly associated with tumour immune-infiltrating cells. In addition, *C3* (23), *CSFR* (24–26), *CXCR6* (27–29), and *CXCL3* (30) have been identified as immune-related factors, and their misexpression in GC affects prognosis (24, 29, 31, 32). Our GO and KEGG analysis results also suggested that these genes are involved in immunoregulation and tumour progression pathways. Then, we used the TCGA-STAD dataset to evaluate the expression of these 28 genes in benign and malignant tissues and their mutations in tumour samples. However, the results of this analysis could not provide valuable insights. Therefore, to further explore the relationship between these 28 genes and the prognosis of GC, we selected five of them to construct a prognostic risk model based on multivariate Cox analysis. Risk prediction models based on polygenic characteristics are commonly used to predict survival outcomes of patients with cancer (33–35). Our prognostic model showed that the expressions of *LAG3* and *CASP1* were negatively correlated with poor prognosis in patients with GC. *LAG3* inhibits the growth of GC and promotes the secretion of CD8+ T cells, *IL-12*, and *IFN- γ* (36), and the expression of *LAG-3* on T-cell surface can be used as a reasonable biomarker of anti-PD-1 therapy (37, 38). In addition, *CASP1* has been shown to be activated by HP infection (39, 40). It has both pro-inflammatory and anti-inflammatory effects because of its different substrates (41). The expression of the other three genes, namely *MAFB*, *DKK3*, and *PDLIM7*, was positively correlated with the poor prognosis of patients with GC. Our results suggested that the risk model based on 28 genes can separate the population and exhibits a superior performance in predicting the prognosis of patients with GC. While analysing the difference in infiltrating immune cells between the high- and low-risk groups, our results suggested that the activated CD4+ T cells and CD8+ T cells, rather than regulatory T cells, are highly enriched in the low-risk groups, which is consistent with the molecular function of *LAG3* and *CASP1*. To understand the underlying mechanism of poor prognosis in high-risk populations, the GSEA was used to identify significantly enriched signalling pathways in the high-risk populations. The results suggested that the high-risk group were enriched in the angiogenesis, hypoxia, macrophage autophagy, and tumour stem cell-related signalling pathway. A report showed that the expression of *MAFB* oncoprotein is regulated by the cytolethal distending toxin of enterohepatic HP (42), and *MAFB* is specifically expressed in tumour-associated macrophages to induce angiogenesis (43). In addition, studies on osteosarcoma have reported that *MAFB* increases the expression of stem cell regulatory factor *SOX9* at the transcriptional level (44). Overall, the activation of carcinogenic pathways induced by misexpression of 28 genes is the cause of poor prognosis in the high-risk group. Here, we identified the first clinical tag of 28 genes: prognostic indicators.

To observe differences among the samples with different modified states of 28 genes, we further divided the patients with GC into two by using the NMF method, namely

cluster_1 and cluster_2. The survival analysis suggested a significant difference in survival between the two groups of patients with GC. The difference analysis showed that cluster_1 with favourable prognosis had a higher expression of immune-related factors, such as *CCL20*, *CXCL2*, and *CXCL3*, and is supported by the signalling pathway analysis. Cytokines, chemokines, and inflammatory response-related signalling pathways were widely enriched in cluster_1. In addition, the enrichment of HP signalling pathway was observed in cluster_1, suggesting that cluster_1 is closer to the state of inflammatory response in the early stage of HP infection. To prove this result, we compared the distribution of infiltrating immune cells between cluster_1 and cluster_2. The results were similar to those observed in the case of high- and low-risk groups. Activated CD4+ T cells, type 17 helper T cells, and neutrophils were highly enriched in cluster_1. This result supported that HP-induced diseases are mainly mediated by Th1 cells and Th1 cytokines (3). In addition, TH17 helper cells fight against the immune response of extracellular bacteria and moulds, and the cytokines released by the helper cells mainly activate neutrophils (45). and are highly consistent with our results. Combined with the aforementioned results, we identified the second clinical tag of 28 genes: HP infection-related feature. Subsequently, we re-verified the difference in the expression of HP prognosis-related genes between cluster_1 and cluster_2, which suggested that *HLA-DMA*, *CASP1*, *CXCR6*, *LAG3*, *VNN2*, and *CXCL3* were highly expressed in cluster_1. *VNN2* is a haematopoietic stem cell marker (46, 47), that participates in inflammation and leukocyte migration (48). However, the role of *VNN2* in GC is unclear. Based on the aforementioned results, we defined these 28 genes as HP-related prognostic genes.

To evaluate the modification patterns of HP-related prognostic genes in a single sample, we established a scoring system based on 28 HP-related prognostic genes and termed it as the HPscore. Comprehensive analysis showed that the HPscore is related to tumour progression and affects tumour prognosis. HP infection leads to the imbalance of DNA methylation in gastric mucosal epithelial cells of the host (49–52). As a result, some proto-oncogenes are activated to induce cancer (53). Microsatellite instability (MSI) in GC also showed specific hypermethylation of DNA (54). Surprisingly, a negative correlation was observed between the HPscore and DNA methylation stemness index and mutation load in TCGA datasets. After optimising the DNA methylation index, the HPscore became unrelated to the DNA methylation level (the results are not shown). In terms of clinical features, MSI was also not related to the HPscore (the results are not provided). The reason may be that in the TCGA-STAD dataset, mDNAsi derived from the one-class logistic regression machine learning algorithm (OCLR) does not sufficiently reflect the methylation level of GC (the high level of tumour cell stemness index in this study is a protective factor for GC prognosis). As reported previously, GC may have multiple stem cell-like genomic characteristics or non-stem phenotypes dominated by hypermethylation (55). Excitingly, we compared the differences in nucleotide_excision_repair, DNA_damage_repair, homologous_recombination, mismatch_repair, and cell_cycle_regulators between the high- and low-score arrays, and the results confirmed our HPscore system. In the high-score group, the ability to repair DNA damage was generally low, suggesting that HP infection impairs the autonomous repair function of cells (56). In addition, the two HPscore groups showed different TME permeation characteristics. The low-score group showed a stronger inflammatory response, whereas the high-score group was accompanied by a large number of stromal cells including fibroblasts and endothelial cells. The subsequent results showed that the enrichment score of CAF markers in the high-HPscore group was higher than that in the low-HPscore group. However, no difference was observed in the HPscores between normal fibroblasts and tumour-associated fibroblasts, suggesting that the modification of HP-related prognostic genes in tumour cells induces the transformation of NF to CAF rather than to fibrous cells. In chronic inflammation and cancer, tissue-resident fibroblasts become the critical cell types that regulate the activation or inhibition of the immune response (11). Fibroblasts assist immune cells to maintain an effective inflammatory environment in chronic inflammation and promote immunosuppression in malignant tumours to assist tumour cells in immune escape (10, 11). In addition, fibroblasts are necessary for the synthesis and remodelling of the extracellular matrix during angiogenesis and germination (57). These new blood vessels bring bone marrow-derived suppressor cells, including BMDCs, into the TME. Chronic HP infection can lead to BMDC recruitment to promote the stemness-like characteristics of GC cells (5, 58). Our results also supported this conclusion. GSEA results suggested that various tumour stem cell-related signalling pathways, such as the Notch signal pathway, Wnt pathway, and Hedgehog pathway, were enriched in the high-score group. BMDCs associated with HP were also significantly enriched in the high-score group. In addition, our study suggested that the Wnt pathway plays a key role in the carcinogenesis induced by HP infection. Studies have reported that HP promotes tumour progression by activating the Wnt/β-catenin pathway (59, 60) and promotes CSC-like characteristics in GC cells (61); these findings are consistent with those of our study. After optimising the tumour stem cell index, we found that the HPscore was positively correlated with the tumour stemness index. Simultaneously, we also proved the positive correlation between the HPscore and EMT and F-TGF-B. These results suggested that HP helps tumour immune escape and angiogenesis by activating fibroblasts and recruits BMDCs to enhance the characteristics of GC stem cells and promote cancer development, in which the Wnt signalling pathway plays a key role.

Combined with the aforementioned evidence, we studied the role of HPscore in treatment. We first evaluated the relationship between the HPscore and PD-L1. Unfortunately, the predictive value of the HPscore in PD1 and anti-PD-L1 immunotherapy is unstable. To date, no detailed report on the relationship between

HP infection and PD1/PD-L1 is available. Because of the complexity of the TME, only a few patients benefit from the treatment of immune checkpoint block (62). Our HPscore may not perform well in diseases that are not related to bacterial infections. Hence, more experiments are needed to verify the interaction between HP and PD-L1. We then predicted the therapeutic efficacy of antimicrobials by using the HPscore. Based on the limited data, we found that the HPscore was positively correlated with the degree of gastritis, which can help predict the grade of gastritis. Metronidazole is used to treat various infectious diseases including HP infection. Studies have reported that the sensitivity to metronidazole decreases in patients with HP infection (63, 64). In the vaginitis data set, the HPscore decreased significantly after three weeks of metronidazole treatment. These results showed that the HPscore plays a guiding role in clinical diagnosis and efficacy evaluation. However, our study has many limitations. First, because of the complexity of HP pathogenesis, the existing HP metadata could not fully reflect the status of HP infection. Second, we found a stronger correlation of HP infection with stromal cells than with the infiltrating immune cells. This result indicated that more communication might exist between stromal cells and HP. Finally, the mechanism through which HP recruits BMDCs remains to elucidated experimentally in detail.

## Conclusions

In conclusion, the HPscore can comprehensively evaluate the permeability characteristics of the individual TME and drug efficacy in patients with GC. In this study, we used HP-related gene datasets to derive the characteristics of HP-related prognostic genes for the first time. Based on the HPscore system, we showed the comprehensive view of the TME of the sample shaped by HP-related prognostic gene modification. Clinically, the HPscore can predict the inflammatory grade of patients with gastritis and reflect the therapeutic effect of metronidazole. Our findings provide a basis and framework for better understanding the carcinogenic mechanism in patients infected with HP and develop an efficient tool for personalised and effective treatment strategies.

## Data availability statement

The datasets presented in this study can be found in online repositories. The names of the repository/repositories and accession number(s) can be found in the article/**Supplementary Material**.

## Ethics statement

The study was granted by the Bioethics Committee of First Affiliated Hospital of Guangxi Medical University (No. 2014-KY-E-006).

## Author contributions

YW and KTZ are responsible for study conceptualization. YW generated most of the data, assisted by KTZ and JCW KTZ and CJW checked the statistical method. YW, KTZ, JCW, and CJW prepared the figures. YW and KTZ wrote the manuscript. JQC provided resources, supervision and editing. All authors contributed to the article and approved the submitted version.

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## Conflict of interest

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

## Publisher's note



## Supplementary material

The Supplementary Material for this article can be found online at: https://www.frontiersin.org/articles/10.3389/fsurg.2022.964203/full#supplementary-material.

# References

1. Smyth E, Nilsson M, Grabsch H, van Grieken N, Lordick F. Gastric cancer. *Lancet (London, England)*. (2020) 396(10251):635–48. doi: 10.1016/S0140-6736(20)31288-5

2. Crowe S. Helicobacter pylori infection. *N Engl J Med*. (2019) 380(12):1158–65. doi: 10.1056/NEJMcp1710945

3. Kusters J, van Vliet A, Kuipers E. Pathogenesis of Helicobacter pylori infection. *Clin Microbiol Rev*. (2006) 19(3):449–90. doi: 10.1128/CMR.00054-05

4. Choi I, Kim C, Lee J, Kim Y, Kook M, Park B, et al. Helicobacter pylori family history of gastric cancer and treatment. *N Engl J Med*. (2020) 382(5):427–36. doi: 10.1056/NEJMoa1909666

5. Bessède E, Dubus P, Mégraud F, Varon C. Helicobacter pylori infection and stem cells at the origin of gastric cancer. *Oncogene*. (2015) 34(20):2547–55. doi: 10.1038/onc.2014.187

6. Houghton J, Stoicov C, Nomura S, Rogers AB, Carlson J, Li H, et al. Gastric cancer originating from bone marrow-derived cells. *Science*. (2004) 306(5701):1568–71. doi: 10.1126/science.1099513

7. Ferrand J, Noel D, Lehours P, Prochazkova-Carlotti M, Chambonnier L, Menard A, et al. Human bone marrow-derived stem cells acquire epithelial characteristics through fusion with gastrointestinal epithelial cells. *PLoS One*. (2011) 6(5):e19569. doi: 10.1371/journal.pone.0019569

8. Ferrand J, Lehours P, Schmid-Alliana A, Mégraud F, Varon C. Helicobacter pylori infection of gastrointestinal epithelial cells in vitro induces mesenchymal stem cell migration through an NF-κB-dependent pathway. *PLoS One*. (2011) 6(12):e29007. doi: 10.1371/journal.pone.0029007

9. Zhang X, Arnold I, Müller A. Mechanisms of persistence, innate immune activation and immunomodulation by the gastric pathogen Helicobacter pylori. *Curr Opin Microbiol*. (2020) 54:1–10. doi: 10.1016/j.mib.2020.01.003

10. Franklin R. Fibroblasts and macrophages: collaborators in tissue homeostasis. *Immunol Rev*. (2021) 302(1):86–103. doi: 10.1111/imr.12989

11. Davidson S, Coles M, Thomas T, Kollias G, Ludewig B, Turley S, et al. Fibroblasts as immune regulators in infection, inflammation and cancer. *Nat Rev Immunol*. (2021) 21(11):704–17. doi: 10.1038/s41577-021-00540-z

12. Sotiriou C, Wirapati P, Loi S, Harris A, Fox S, Smeds J, et al. Gene expression profiling in breast cancer: understanding the molecular basis of histologic grade to improve prognosis. *J Natl Cancer Inst*. (2006) 98(4):262–72. doi: 10.1093/jnci/djj052

13. Zeng D, Li M, Zhou R, Zhang J, Sun H, Shi M, et al. Tumor microenvironment characterization in gastric cancer identifies prognostic and immunotherapeutically relevant gene signatures. *Cancer Immunol Res*. (2019) 7(5):737–50. doi: 10.1158/2326-6066.CIR-18-0436

14. Yoshihara K, Shahmoradgoli M, Martinez E, Vegesna R, Kim H, Torres-Garcia W, et al. Inferring tumour purity and stromal and immune cell admixture from expression data. *Nat Commun*. (2013) 4:2612. doi: 10.1038/ncomms3612

15. Charoentong P, Finotello F, Angelova M, Mayer C, Efremova M, Rieder D, et al. Pan-cancer immunogenomic analyses reveal genotype-immunophenotype relationships and predictors of response to checkpoint blockade. *Cell Rep*. (2017) 18(1):248–62. doi: 10.1016/j.celrep.2016.12.019

16. Chen DS, Mellman I. Elements of cancer immunity and the cancer-immune set point. *Nature*. (2017) 541(7637):321–30. doi: 10.1038/nature21349

17. Nguyen T, Kim S, Park J, Hahm K, Lee H. Repressed TGF-β signaling through CagA-Smad3 interaction as pathogenic mechanisms of Helicobacter pylori-associated gastritis. *J Clin Biochem Nutr*. (2015) 57(2):113–20. doi: 10.3164/jcbn.15-38

18. El-Zaatari M, Bishu S, Zhang M, Grasberger H, Hou G, Haley H, et al. Aim2-mediated/IFN-β-independent regulation of gastric metaplastic lesions via CD8+ T cells. *JCI Insight*. (2020) 5(5):e94035. doi: 10.1172/jci.insight.94035

19. Mueller S, Mackay L. Tissue-resident memory T cells: local specialists in immune defence. *Nat Rev Immunol*. (2016) 16(2):79–89. doi: 10.1038/nri.2015.3

20. Park C, Kupper T. The emerging role of resident memory T cells in protective immunity and inflammatory disease. *Nat Med*. (2015) 21(7):688–97. doi: 10.1038/nm.3883

21. Song S, Min H, Niu M, Wang L, Wu Y, Zhang B, et al. S1PR1 predicts patient survival and promotes chemotherapy drug resistance in gastric cancer cells through STAT3 constitutive activation. *EBioMedicine*. (2018) 37:168–76. doi: 10.1016/j.ebiom.2018.10.005

22. Yeon M, Kim Y, Pathak D, Kwon E, Kim D, Jeong M, et al. The CAGE-MiR-181b-5p-S1PR1 axis regulates anticancer drug resistance and autophagy in gastric cancer cells. *Front Cell Dev Biol*. (2021) 9:666387. doi: 10.3389/fcell.2021.666387

23. Yuan K, Ye J, Liu Z, Ren Y, He W, Xu J, et al. Complement C3 overexpression activates JAK2/STAT3 pathway and correlates with gastric cancer progression. *J Exp Clin Cancer Res*. (2020) 39(1):9. doi: 10.1186/s13046-019-1514-3

24. Chen D, Xiong L, Zhang L, Yu H, Xu Y, Wang M, et al. CSF1R is a prognostic biomarker and correlated with immune cell infiltration in the gastric cancer microenvironment. *Pharmgenomics Pers Med*. (2021) 14:445–57. doi: 10.2147/PGPM.S301303

25. Huo J, Wu L, Zang Y. Construction and validation of a universal applicable prognostic signature for gastric cancer based on seven immune-related gene correlated with tumor associated macrophages. *Front Oncol*. (2021) 11:635324. doi: 10.3389/fonc.2021.635324

26. Okugawa Y, Toiyama Y, Ichikawa T, Kawamura M, Yasuda H, Fujikawa H, et al. Colony-stimulating factor-1 and colony-stimulating factor-1 receptor co-expression is associated with disease progression in gastric cancer. *Int J Oncol*. (2018) 53(2):737–49. doi: 10.3892/ijo.2018.4406

27. Deutsch A, Steinbauer E, Hofmann N, Strunk D, Gerlza T, Beham-Schmid C, et al. Chemokine receptors in gastric MALT lymphoma: loss of CXCR4 and upregulation of CXCR7 is associated with progression to diffuse large B-cell lymphoma. *Mod Pathol*. (2013) 26(2):182–94. doi: 10.1038/modpathol.2012.134

28. Han J, Fu R, Chen C, Cheng X, Guo T, Huangfu L, et al. CXCL16 promotes gastric cancer tumorigenesis via ADAM10-dependent CXCL16/CXCR6 axis and activates Akt and MAPK signaling pathways. *Int J Biol Sci*. (2021) 17(11):2841–52. doi: 10.7150/ijbs.57826

29. Jin J, Dai F, Long Z, Cai H, Liu X, Zhou Y, et al. CXCR6 Predicts poor prognosis in gastric cancer and promotes tumor metastasis through epithelial-mesenchymal transition. *Oncol Rep*. (2017) 37(6):3279–86. doi: 10.3892/or.2017.5598

30. Yamamoto Y, Kuroda K, Sera T, Sugimoto A, Kushiyama S, Nishimura S, et al. The clinicopathological significance of the CXCR2 ligands, CXCL1, CXCL2, CXCL3, CXCL5, CXCL6, CXCL7, and CXCL8 in gastric cancer. *Anticancer Res*. (2019) 39(12):6645–52. doi: 10.21873/anticanres.13879

31. Wu T, Wang C, Wang W, Liu L, Yun J, Zhou Z. Association of preoperative and postoperative CA72-4 with gastric cancer outcome. *J Surg Oncol*. (2021) 123(8):1699–707. doi: 10.1002/jso.26446

32. Ye J, Ren Y, Chen J, Song W, Chen C, Cai S, et al. Prognostic significance of preoperative and postoperative complement C3 depletion in gastric cancer: a three-year survival investigation. *BioMed Res Int*. (2017) 2017:2161840. doi: 10.1155/2017/2161840

33. Chen H, Yu S, Chen C, Chang G, Chen C, Yuan A, et al. A five-gene signature and clinical outcome in non-small-cell lung cancer. *N Engl J Med*. (2007) 356(1):11–20. doi: 10.1056/NEJMoa060096

34. Low Y, Blöcker C, McPherson J, Tang S, Cheng Y, Wong J, et al. A formalin-fixed paraffin-embedded (FFPE)-based prognostic signature to predict metastasis in clinically low risk stage I/II microsatellite stable colorectal cancer. *Cancer Lett*. (2017) 403:13–20. doi: 10.1016/j.canlet.2017.05.031

35. Zhang J, Song W, Chen Z, Wei J, Liao Y, Lei J, et al. Prognostic and predictive value of a microRNA signature in stage II colon cancer: a microRNA expression analysis. *Lancet Oncol*. (2013) 14(13):1295–306. doi: 10.1016/S1470-2045(13)70491-1

36. Li N, Jilisihan B, Wang W, Tang Y, Keyoumu S. Soluble LAG3 acts as a potential prognostic marker of gastric cancer and its positive correlation with CD8+T cell frequency and secretion of IL-12 and INF-γ in peripheral blood. *Cancer Biomark*. (2018) 23(3):341–51. doi: 10.3233/CBM-181278

37. Park Y, Seo A, Koh J, Nam S, Kwak Y, Ahn S, et al. Expression of the immune checkpoint receptors PD-1, LAG3, and TIM3 in the immune context of stage II and III gastric cancer by using single and chromogenic multiplex immunohistochemistry. *Oncoimmunology*. (2021) 10(1):1954761. doi: 10.1080/2162402X.2021.1954761

38. Ohmura H, Yamaguchi K, Hanamura F, Ito M, Makiyama A, Uchino K, et al. OX40 And LAG3 are associated with better prognosis in advanced gastric cancer patients treated with anti-programmed death-1 antibody. *Br J Cancer*. (2020) 122(10):1507–17. doi: 10.1038/s41416-020-0810-1

39. Koch K, Hartung M, Urban S, Kyburz A, Bahlmann A, Lind J, et al. Helicobacter urease-induced activation of the TLR2/NLRP3/IL-18 axis protects against asthma. *J Clin Invest*. (2015) 125(8):3297–302. doi: 10.1172/JCI79337

40. Ng G, Menheniott T, Every A, Stent A, Judd L, Chionh Y, et al. The MUC1 mucin protects against Helicobacter pylori pathogenesis in mice by regulation of the NLRP3 inflammasome. *Gut*. (2016) 65(7):1087–99. doi: 10.1136/gutjnl-2014-307175

41. Hitzler I, Sayi A, Kohler E, Engler D, Koch K, Hardt W, et al. Caspase-1 has both proinflammatory and regulatory properties in Helicobacter infections, which are differentially mediated by its substrates IL-1β and IL-18. *J Immunol*. (2012) 188(8):3594–602. doi: 10.4049/jimmunol.1103212

42. Péré-Védrenne C, He W, Azzi-Martin L, Prouzet-Mauléon V, Buissonnière A, Cardinaud B, et al. The nuclear remodeling induced by Helicobacter cytolethal distending toxin involves MAFB oncoprotein. *Toxins (Basel)*. (2020) 12(3):174. doi: 10.3390/toxins12030174

43. Yadav M, Inoue Y, Nakane-Otani A, Tsunakawa Y, Jeon H, Samir O, et al. Transcription factor MafB is a marker of tumor-associated macrophages in both mouse and humans. *Biochem Biophys Res Commun*. (2020) 521(3):590–5. doi: 10.1016/j.bbrc.2019.10.125

44. Chen Y, Wang T, Huang M, Liu Q, Hu C, Wang B, et al. MAFB promotes cancer stemness and tumorigenesis in osteosarcoma through a Sox9-mediated positive feedback loop. *Cancer Res*. (2020) 80(12):2472–83. doi: 10.1158/0008-5472.CAN-19-1764

45. Evans R, Antonelou M, Sathiananthamoorthy S, Rega M, Henderson S, Ceron-Gutierrez L, et al. Inherited salt-losing tubulopathies are associated with immunodeficiency due to impaired IL-17 responses. *Nat Commun*. (2020) 11 (1):4368. doi: 10.1038/s41467-020-18184-3

46. Bornhauser B, Cario G, Rinaldi A, Risch T, Rodriguez Martinez V, Schütte M, et al. The hematopoietic stem cell marker VNN2 is associated with chemoresistance in pediatric B-cell precursor ALL. *Blood Adv*. (2020) 4 (17):4052–64. doi: 10.1182/bloodadvances.2019000938

47. Soler D, Young A, Cooper K, Kerstetter-Fogle A, Barnholtz-Sloan J, Gittleman H, et al. The ratio of HLA-DR and VNN2 expression on CD14 myeloid derived suppressor cells can distinguish glioblastoma from radiation necrosis patients. *J Neuro-Oncol*. (2017) 134(1):189–96. doi: 10.1007/s11060-017-2508-7

48. Sayasith K, Sirois J, Lussier J. Expression, regulation, and promoter activation of vanin-2 (VNN2) in bovine follicles prior to ovulation. *Biol Reprod*. (2013) 89(4):98. doi: 10.1095/biolreprod.113.111849

49. Maeda M, Moro H, Ushijima T. Mechanisms for the induction of gastric cancer by Helicobacter pylori infection: aberrant DNA methylation pathway. *Gastric Cancer*. (2017) 20(Suppl 1):8–15. doi: 10.1007/s10120-016-0650-0

50. Matsusaka K, Funata S, Fukayama M, Kaneda A. DNA Methylation in gastric cancer, related to Helicobacter pylori and Epstein-Barr virus. *World J Gastroenterol*. (2014) 20(14):3916–26. doi: 10.3748/wjg.v20.i14.3916

51. Shin CM, Kim N, Lee HS, Park JH, Ahn S, Kang GH, et al. Changes in aberrant DNA methylation after Helicobacter pylori eradication: a long-term follow-up study. *Int J Cancer*. (2013) 133(9):2034–42. doi: 10.1002/ijc.28219

52. Tahara S, Tahara T, Horiguchi N, Kato T, Shinkai Y, Yamashita H, et al. DNA Methylation accumulation in gastric mucosa adjacent to cancer after Helicobacter pylori eradication. *Int J Cancer*. (2019) 144(1):80–8. doi: 10.1002/ijc.31667

53. Huang KK, Ramnarayanan K, Zhu F, Srivastava S, Xu C, Tan ALK, et al. Genomic and epigenomic profiling of high-risk intestinal metaplasia reveals molecular determinants of progression to gastric cancer. *Cancer Cell*. (2018) 33 (1):137–50. e5. doi: 10.1016/j.ccell.2017.11.018

54. Usui G, Matsusaka K, Mano Y, Urabe M, Funata S, Fukayama M, et al. DNA methylation and genetic aberrations in gastric cancer. *Digestion*. (2021) 102 (1):25–32. doi: 10.1159/000511243

55. Malta TM, Sokolov A, Gentles AJ, Burzykowski T, Poisson L, Weinstein JN, et al. Machine learning identifies stemness features associated with oncogenic dedifferentiation. *Cell*. (2018) 173(2):338–54.e15. doi: 10.1016/j.cell.2018.03.034

56. Sierra J, Piazuelo M, Luis P, Barry D, Allaman M, Asim M, et al. Spermine oxidase mediates Helicobacter pylori-induced gastric inflammation, DNA damage, and carcinogenic signaling. *Oncogene*. (2020) 39(22):4465–74. doi: 10.1038/s41388-020-1304-6

57. Hultgren N, Fang J, Ziegler M, Ramirez R, Phan D, Hatch M, et al. Slug regulates the Dll4-Notch-VEGFR2 axis to control endothelial cell activation and angiogenesis. *Nat Commun*. (2020) 11(1):5400. doi: 10.1038/s41467-020-18633-z

58. Varon C, Dubus P, Mazurier F, Asencio C, Chambonnier L, Ferrand J, et al. Helicobacter pylori infection recruits bone marrow-derived cells that participate in gastric preneoplasia in mice. *Gastroenterology*. (2012) 142(2):281–91. doi: 10.1053/j.gastro.2011.10.036

59. Liu N, Zhou N, Chai N, Liu X, Jiang H, Wu Q, et al. Helicobacter pylori promotes angiogenesis depending on Wnt/beta-catenin-mediated vascular endothelial growth factor via the cyclooxygenase-2 pathway in gastric cancer. *BMC cancer*. (2016) 16:321. doi: 10.1186/s12885-016-2351-9

60. Meng L, Shi H, Wang Z, Fan M, Pang S, Lin R. The gamma-glutamyltransferase gene of Helicobacter pylori can promote gastric carcinogenesis by activating Wnt signal pathway through up-regulating TET1. *Life Sci*. (2021) 267:118921. doi: 10.1016/j.lfs.2020.118921

61. Yong X, Tang B, Xiao Y, Xie R, Qin Y, Luo G, et al. Helicobacter pylori upregulates Nanog and Oct4 via Wnt/β-catenin signaling pathway to promote cancer stem cell-like properties in human gastric cancer. *Cancer Lett*. (2016) 374(2):292–303. doi: 10.1016/j.canlet.2016.02.032

62. Patel S, Minn A. Combination cancer therapy with immune checkpoint blockade: mechanisms and strategies. *Immunity*. (2018) 48(3):417–33. doi: 10.1016/j.immuni.2018.03.007

63. Lee G, Lee K, Shin S, Kang J, Noh C, Kim J, et al. Impact of previous metronidazole exposure on metronidazole-based second-line quadruple therapy for Helicobacter pylori infection. *Korean J Intern Med*. (2020) 35(5):1094–103. doi: 10.3904/kjim.2020.174

64. Regnath T, Raecke O, Enninger A, Ignatius R. Increasing metronidazole and rifampicin resistance of Helicobacter pylori isolates obtained from children and adolescents between 2002 and 2015 in southwest Germany. *Helicobacter*. (2017) 22(1):e12327. doi: 10.1111/hel.12327

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# Comparative study on the clinical effect of preparing neobladder with different lengths of ileum

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**Objective:** To investigate the relationship between orthotopic U-shaped ileal neobladder volume and bladder function. To investigate the correlation between the volume of the radical cystectomy and the U-shaped ileal neobladder *in situ* and the function of the bladder.

**Methods:** The clinical data of patients undergoing in orthotopic U-shaped neobladder in our hospital were retrospectively analyzed. They were divided into two groups according to the length of the retained ileum. Group 1: The length of the ileum was 25-35cm (including 35cm), and the second group: the length of the ileum was 35-45cm. The basic information, cushion usage, urodynamic examination and complications of the two groups were obtained.

**Results:** A total of 88 patients were included in the study, including 33 in the first group and 55 in the second group. There was no statistical difference in general data, lymph node collection, lymph node positive rate, positive margin rate, postoperative pathological stage, pathological grade, pathological type, intraoperative blood loss, blood transfusion, postoperative hospital stay, and complications between the two groups of patients. significance. Although the usage of urine pads in group 1 was more than that in group 2 in the short term after operation ($P<0.05$), it started from the third year after operation. Patients in group 1 used less cushion than group 2 ($P<0.05$). Urodynamic examination was performed on the patients, and the bladder function of group 1 maintained satisfactory time longer than that of group 2. The total number of deaths in the two groups was 12 and 23, respectively. The 5-year overall survival (OS) rate of group 1 was 53.60%, and the 5-year overall survival rate of group 2 was 52.9%.

**Conclusions:** A new bladder formed by cutting the ileum with a length of 25-35 cm (including 35cm) has a longer time to maintain good bladder function than cutting the ileum with a length of 35-45 cm to produce a new bladder.

KEYWORDS

**bladder tumor, radical cystectomy, ileal neobladder surgery, bladder volume, bladder function**

## Introduction

Currently, urinary reconstruction is divided into two categories: incontinence diversions, including ileal conduit (Bricker) diversion, and continental diversions, such as skin reservoirs and orthotopic neobladder connected to the urethra (1). In orthotopic neobladder surgery is the closest to a normal bladder in terms of anatomy and function, does not require an abdominal wall stoma, maintains a good personal image, and improves the quality of life. It has been popularized and applied in recent years (2, 3). Urinary control after neobladder surgery is mainly affected by the anatomical preservation of intraoperative neurovascular and sphincter. We retrospectively analyzed the clinical data of 88 patients with locally advanced bladder cancer in our hospital from May 2014 to May 2019. The patients were divided into two groups according to the length of the ileum intercepted during the operation, thereby affecting the size of the bladder capacity. The length of the intercepted ileum in one group was 25-35cm, and the length of the intercepted ileum in the second group was 35-45cm. To explore the correlation between different bladder capacity and bladder function.

## Materials and methods

This study reviewed 88 cases of locally advanced bladder cancer (cT2-3) admitted in our hospital from May 2014 to May 2019. The inclusion criteria included patients with better urinary control through preoperative urodynamic examination. Patients complete a follow-up for 2-7 years after surgery. The patients were divided into two parallel observation groups according to the length of the ileum intercepted during the operation. Observation group one: 33 cases, 30 males and 3 females; age from 48 to 77 years old with an average age of 60 years; Observation group 2: A total of 55 cases, 50 males and 5 females; aged 46-82 years old, with an average age of 64 years old; the two groups of patients have different ileum interception lengths. Observation group one: the length of the ileum cut was 25-35cm; the observation group two: the length of the cut ileum was 35-45cm.

The operations were performed by the same team of experienced doctors. Both groups of patients were under general anesthesia, and the patients were in a supine position. After entering the pelvic cavity through an abdominal incision, they underwent lymph node dissection and cystectomy. Separate a section of ileum from the ileocecal area, and perform a U-fold folding with the extremities facing the patient's head. A mechanical stapler is used for lateral intestinal-intestinal anastomosis to establish intestinal continuity. The separated intestinal segment is folded in a U-shape, and the jaws of the mechanical stapler are placed into the intestinal segment opening for cutting and anastomosis. In order to complete the pouch to the tube, an opening is made at the lowest point of the U-shaped ileum, and the jaws of the mechanical stapler pass through the opening to complete the U-bag.

We use the cutting closure device to make the U-shaped neobladder of the ileum. This method reduces the length of the intestine required and reduces the tension between the urethra and the anastomosis of the neobladder. The neurovascular bundle is preserved during the operation. After the operation, a new bladder is flushed through a catheter to dilute the ileal mucus and prevent the catheter from being blocked. Postoperative angiography was performed to evaluate reflux and leakage.

We collected and evaluated the basic data of the two groups of patients, the clinical stage of the tumor, the volume of the new bladder, the residual urine volume, the maximum urine flow rate, the recovery of postoperative urinary control, and postoperative complications. The standard for good urinary control after surgery: the number of urine pads used is less than or equal to 1 piece (4), and the number of urine pads used for poor urinary control is> 1 piece. Detect maximum bladder volume, residual urine volume, maximum urine flow rate, etc. according to the technical report standards of the International Association of Urological Control. The bladder function of the two groups was compared through regular outpatient check-ups and telephone follow-up.

Postoperative follow-up was conducted in the form of telephone contact and outpatient follow-up, with an interval of 3 months in the first year, 6 months in the second year, and annual follow-up. The bladder function training after the operation is carried out about 1 week after the operation, and the urinary catheter is clamped and opened regularly. The time starts from every half hour and gradually opens once every 2 to 3 hours. The urine output varies from low to high, until about 250 mL of urine is excreted from the catheter each time. The urination habit training is carried out after the catheter is removed. According to the patient's living habits and activity requirements, a urination plan is formulated. Generally, the patient is instructed to urinate 6 to 8 times during the day and 2 to 3 times at night. Voiding pattern training: They are taught to empty the neobladder by increasing intra-abdominal pressure and relaxing the pelvic floor. Urinary continence training: by repeatedly contracting and relaxing the pelvic floor muscles, to restore urinary continence as soon as possible and eliminate urinary incontinence.

The SPSS 25.0 software was used to statistically process the data, and the Kruskal-Wallis, T test and X2 test were used to

analyze the data. The difference was statistically significant with P<0.05.

## Results

We evaluated 88 patients who underwent surgery. The median follow-up time of 33 patients in observation group 1 was 44 months (27-65 months), and the median follow-up time of 55 patients in observation group 2 was 65 months (42-82 months). There was no significant difference in the follow-up time between the two groups (P>0.05). The BMI of most patients was within the normal range, and the difference between the two groups was not statistically significant. There was no significant difference in the clinical staging of tumors between the two groups. There was also no significant difference in diabetes and postoperative pathological lymph node positive between the two groups (Table S1).

There was no significant difference in preoperative hematological indexes (including Cr, BUN, HP) between the two groups. The operation time of group 1 was slightly shorter than that of group 2, but the difference was not statistically significant. There was no significant difference in estimated intraoperative blood loss between the two groups (344.0 ± 159.0 *VS* 359 ± 163.1 P=0.629). The number of intraoperative or postoperative blood transfusions in group 2 was more than that in group 1, but there was no significant difference in blood transfusion between the two groups. The postoperative hospital stay was 16.0 ± 3.3 *VS* 17.8 ± 4.4 in the two groups respectively, and the difference was not statistically significant. There was no significant difference in early postoperative complications and late complications (Table S2)

The results showed that in our perioperative complications study, the electrolyte disturbance in group 1 was less than that in group 2 (P <0.05), and the incidence of hydronephrosis in group 2 was significantly higher than that in group 1. The time of exhaust and defecation in group 1 was significantly earlier than that in group 2, and there was no significant difference in other complications between the two groups. (Table 1).

The usage of the changing pad reflects the bladder function of the patient. One year after the operation, the daily usage of urine pad in observation group 2 was less than that in observation group 1. At the same time, more than half of the patients have good urinary control ability during the day and only need to use the urine pad at night. With the prolongation of monitoring time, the use of urine pads in both groups improved significantly. By the second year after surgery, there was no statistically significant difference in the use of changing pads and the use of changing pads during the day and night between the two groups (P>0.05). Further follow-up, regardless of the comparison of the usage of the urine pad and the usage, the observation group 1 was significantly better than the observation group 2 (Table 2).

The use of changing pads reflects the patient's ability to control urine. At 1 year after surgery, the daily usage of changing pads in group 2 was less than that in observation group 1. At the same time, more than half of the patients had good urination control during the day, and only needed to use a urine pad at night. With the extension of follow-up time, the use of urine pads in both groups was significantly improved. By the 2nd year after operation, the patients in the two groups were better than the patients in the first group in terms of the number of urine pads used and the use of the white night pads. With the extension of follow-up time, the number of patients in group 1 gradually decreased, while the use of pads in group 2 increased gradually after the third year of follow-up (Table 3).

The size and wetness of the pad are important indicators of urinary incontinence. In our study, the size of day and night pad

TABLE 1 Perioperative and postoperative information of the two groups.

| Variable | Group1 | Group2 | Pvalue |
|---|---|---|---|
| Perioperative complications | | | |
| Infection | 7 | 12 | 0.947 |
| Gastrointestinal tract related | 3 | 8 | 0.677 |
| Urinary fistula | 2 | 2 | 0.629 |
| Disturbance of electrolyte | 9 | 27 | 0.044 |
| Postoperative complications | | | |
| Infection | 5 | 10 | 0.714 |
| Chronic pyelonephritis | 3 | 10 | 0.394 |
| Kidney seeper | 3 | 17 | 0.036 |
| Bladder calculi | 5 | 10 | 0.714 |
| Gastrointestinal tract related | 3 | 7 | 0.862 |
| Time to flatus | 35.5 ± 16.5 | 44.5 ± 17.2 | 0.013 |
| Time to bowel | 116.1 ± 21.3 | 125.7 ± 23.9 | 0.045 |

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TABLE 2 Postoperative pathological results of bladder cancer patients in two groups.

| Variable | Group 1 | Group 2 | P value |
|---|---|---|---|
| Surgical margins | | | 0.629 |
| Negative | 31 | 53 | |
| Positive | 2 | 2 | |
| Number of lymph nodes retrieved | 18.0 ± 2.8 | 19.2 ± 3.5 | 0.099 |
| Positive lymph node | 2 | 2 | 0.629 |
| Pathological stage | | | 0.984 |
| Tis | 3 | 6 | |
| T1 | 19 | 32 | |
| T2 | 7 | 12 | |
| T3 | 4 | 5 | |
| Tumor grade | | | 0.607 |
| G1 | 6 | 15 | |
| G2 | 17 | 24 | |
| G3 | 10 | 16 | |
| Pathological type | | | 0.333 |
| Transitional cell carcinoma | 31 | 50 | |
| Squamous cell carcinoma | 2 | 3 | |
| Adenocarcinoma | 1 | 2 | |

use in group 1 was significantly smaller than that in group 2. It was found that there was no significant difference between group 1 and group 2 in the degree of pad wetting during the day (P =0.073). However, the pads were significantly wetter at night in group 2 than in group 1 (Table 4).

Within 1 year after the completion of radical bladder resection and orthotopic U-shaped neobladder, there was no significant difference in residual urine between the two groups (P>0.05). Subsequently, the bladder residual urine in observation group 1 was significantly less than that in observation group 2, and the difference in average residual urine volume between the two groups gradually increased. The residual urine volume of observation group 2 first showed a downward trend, and gradually increased in the 4th year after surgery. The maximum urine flow rate can reflect bladder function. The average maximum urine flow rate of observation group 1 after operation showed an upward trend. By the fifth year after operation, the average maximum urine flow rate was 19.0 ± 2.3 mL/s. However, the peak of the maximum urine flow rate in observation group 2 was in the 4th year after surgery. Too large or too small a new bladder will affect the function of the bladder. The longer it stays within a certain range, the more beneficial it will be to the patient's postoperative urine control. Due to the different length of the intercepted ileum between the two groups of patients, the maximum bladder volume after the operation of the two groups has always been different (Table 5).

The follow-up time of the patients in the two groups was 43.8 ± 13.0 and 47.5 ± 15.7 months, respectively, and the overall deaths were 12 and 23, respectively. The 5-year overall survival (OS)in group 1 was 53.60%, and the 5-year OS in group 2 was 52.90% (Figure 1). There was no significant difference in OS between the two groups.(P = 0.657; HR = 0.855; 95CI = 0.429 to 1.705).

## Discussion

Among urinary system tumors, the incidence or mortality of bladder cancer is extremely high. Radical cystectomy (RC) + urinary diversion (UD) is the gold standard for the treatment of muscular invasive bladder cancer and high-risk non-muscular invasive bladder cancer (5). As the age of onset of bladder cancer tends to be younger and medical conditions improve, many patients still have strong social requirements at the time of onset. RC+traditional urinary diversion surgery requires an abdominal wall stoma and an external urine bag, which affects the quality of life of patients after surgery. With the rise of the concept of urinary system reconstruction, especially the development of "in orthotopic ileal neobladder" surgery, bladder orthotopic reconstruction as a controllable urinary diversion surgery is gradually being carried out in various medical centers. However, this procedure takes a long time and the surgical technique is difficult, leading to its slow development. For this reason, urologists are actively exploring bladder reconstruction techniques in order to build an "ideal" new bladder with a shorter operation time and smaller abdominal incisions.

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TABLE 3 The use of changing pads by patients.

| Variable | Group 1 | Group 2 | P value |
|---|---|---|---|
| One year after surgery | | | |
| Pads per 24 h | | | 0.032 |
| 0-1 | 17/33 (51.5) | 40/54 (74.1) | |
| ≥2 | 16/33 (48.5) | 14/54 (25.9) | |
| Pad use | | | 0.017 |
| Day only | 0/33 (0) | 0 (0) | |
| Night only | 17/33 (51.5) | 41/54 (75.9) | |
| Day and night | 16/33 (48.5) | 13/54 (24.1) | |
| Two years after surgery | | | |
| Pads per 24 h | | | 0.351 |
| 0-1 | 22/31 (71.0) | 40/50 (80.0) | |
| ≥2 | 9/31 (29.0) | 10/50 (20.0) | |
| Pad use | | | 0.559 |
| Day only | 0 (0) | 0 (0) | |
| Night only | 24/31 (77.4) | 42/50 (84.0) | |
| Day and night | 7/31 (22.6) | 8/50 (16.0) | |
| Three years after surgery | | | |
| Pads per 24 h | | | 0.032 |
| 0-1 | 20/24 (83.3) | 26/44 (59.1) | |
| ≥2 | 4/24 (16.7) | 18/44 (40.9) | |
| Pad use | | | 0.367 |
| Day only | 1/24 (4.2) | 2/44 (4.5) | |
| Night only | 17/24 (70.8) | 27/44 (61.4) | |
| Day and night | 4/24 (16.7) | 14/44 (31.8) | |
| No | 2/24 (8.3) | 1/44 (2.3) | |
| Four years after surgery | | | |
| Pads per 24 h | | | 0.040 |
| 0-1 | 12/14 (85.7) | 16/33 (48.5) | |
| ≥2 | 2/14 (14.3) | 17/33 (34.8) | |
| Pad use | | | 0.004 |
| Day only | 1/14 (7.1) | 1/33 (3.0) | |
| Night only | 8/14 (57.1) | 13/33 (39.4) | |
| Day and night | 2/14 (14.3) | 19/33 (57.6) | |
| No | 3/14 (21.4) | 0/33 (0) | |
| Five years after surgery | | | |
| Pads per 24 h | | | 0.021 |
| 0-1 | 6/7 (85.7) | 8/21 (38.1) | |
| ≥2 | 1/7 (15.3) | 13/21 (61.9) | |
| Pad use | | | 0.048 |
| Day only | 0 (0) | 0 (0) | |
| Night only | 5/7 (71.4) | 6/21 (28.6) | |
| Day and night | 1/7 (14.3) | 15/21 (71.4) | |
| No | 1/7 (14.3) | 0 | |

Experience has shown that non-absorbable titanium nails have been safely used in various urology laparotomy and laparoscopic removal and reconstruction operations, including bladder cuff resection in nephroureterectomy. Based on this, Abreu (4) and others reported their ileal neobladder operation, which has the characteristics of "simple, fast and effective". Using a mechanical stapler can quickly establish a new ileal bladder and significantly reduce the operation time. Inspired by this research, we carried

TABLE 4 Patterns of mucus leakage for two groups.

| Variable | Group1 | Group2 | P value |
|---|---|---|---|
| Pad size(daytime) | | | <0.00 |
| No use | 6/7 | 6/21 | |
| Small | 0/7 | 1/21 | |
| medium | 1/7 | 8/21 | |
| Large | 0/7 | 6/21 | |
| Pad size(nighttime) | | | 0.042 |
| No use | 1/7 | 0/7 | |
| Small | 3/7 | 4/21 | |
| medium | 1/7 | 14/21 | |
| Large | 2/7 | 3/21 | |
| Pad wetness(daytime) | | | 0.073 |
| No use | 6/7 | 6/21 | |
| Almost dry | 0/7 | 1/21 | |
| Slightly wet | 1/7 | 3/21 | |
| Wet | 0/7 | 3/21 | |
| Soaked | 0/7 | 8/21 | |
| Pad wetness(nighttime) | | | 0.025 |
| No use | 1/7 | 0/7 | |
| Almost dry | 1/7 | 2/21 | |
| Slightly wet | 4/7 | 3/21 | |
| Wet | 1/7 | 10/21 | |
| Soaked | 0/7 | 6/21 | |

out laparoscopic use of mechanical stapler and closing device to establish ileal neobladder.

There are some technical difficulties in the operation of RARC combined with urinary diversion. Previous experience shows that RARC is feasible to combine with urinary diversion, and is superior to LRC and ORC in many aspects (including Urinary function) (6–8). The team has rich LRC/RARC operation experience. In our previous research, we found that both RARC and LRC are safe and effective (9).

Initially, we used a 35-45 cm ileum for neobladder preparation during laparoscopic in orthotopic U-shaped ileal neobladder surgery. During the postoperative follow-up of the patient, it was found that the bladder function did not meet our ideal expectations. After analysis, it was found that the new bladder capacity expanded too quickly with time, and the time to maintain good bladder function was too short. To this end, we actively explored a shorter ileum for neobladder preparation, and finally decided to use a 25-35cm length of ileum for neobladder preparation.

After orthotopic bladder replacement, the most noteworthy issues include urinary tract changes and urinary dysfunction. As urine fills the new bladder, it acts as a low-pressure reservoir. During urination, the pressure on the abdomen, renal pelvis, and bladder increase at the same time, and the use of Valsalva will promote the emptying of urine without reflux (10, 11). In addition, the unidirectional peristalsis of the ureter and proximal segment of the ileum acts as a dynamic anti-reflux system during the filling phase (12).

In this study, 88 patients who used bladder substitutes were followed up for at least 2 years. In the comparison of the rate of daytime urinary incontinence, the incontinence rate of observation group 1 was higher than that of observation group 2 in the first year after operation (36.4% *VS* 23.6%); by the second year after operation, the difference in the rate of day incontinence between the two groups decreased (24.2% *VS* 16.4%); from the third year after the operation, the day incontinence rate of observation group 1 was always lower than that of observation group 2 during the follow-up period: (3rd year after operation: 8.0% *VS* 9.1%; fourth year after operation: 5.9% *VS* 13.0%; the fifth year after surgery: 0% *VS* 34.3%);. The nocturnal incontinence rate of the two groups also showed the above trend, (the first year after surgery: 42.4% *VS* 27.3%; the second year after surgery: 27.3% *VS* 25.5%; the third year after surgery: 20.0% *VS* 23.6%; the first year after surgery: 20.0% *VS* 23.6%; Four years: 11.8% *VS* 34.8%; fifth year after surgery: 14.3% *VS* 37.1%). According to reports, the urinary control rate after Intracorporeal orthotopic neobladder is 80-100% during the day and 45-90% at night (13–15).

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TABLE 5 Patient's urodynamic parameters.

| Variable | Group 1 | Group 1 | P value |
|---|---|---|---|
| One month after surgery | n=33 | n=55 | |
| Residual urine | 66.4 ± 16.4 | 75.8 ± 19.4 | 0.296 |
| Maximum flow rate (mL/sec) | 8.8 ± 2.1 | 9.2 ± 1.8 | 0.169 |
| Maximum reservoir capacity (mL) | 232.6 ± 27.0 | 312.4 ± 24.4 | 0.000 |
| Six months after surgery | n=33 | n=55 | |
| Residual urine | 68.5 ± 12.3 | 67.5 ± 17.5 | 0.581 |
| Maximum flow rate (mL/sec) | 10.3 ± 2.4 | 12.4 ± 2.1 | 0.542 |
| Maximum reservoir capacity (mL) | 305.0 ± 28.3 | 387.1 ± 27.1 | 0.000 |
| One year after surgery | n=33 | n=54 | |
| Residual urine | 55.0 ± 14.5 | 58.3 ± 25.2 | 0.826 |
| Maximum flow rate (mL/sec) | 12.9 ± 2.8 | 13.1 ± 1.2 | 0.740 |
| Maximum reservoir capacity (mL) | 361.0 ± 33.2 | 440.1 ± 30.1 | 0.000 |
| Two years after surgery | n=31 | n=50 | |
| Residual urine | 42.8 ± 17.1 | 54.1 ± 44.3 | 0.038 |
| Maximum flow rate (mL/sec) | 15.8 ± 3.6 | 15.2 ± 3.5 | 0.310 |
| Maximum reservoir capacity (mL) | 390.1 ± 31.1 | 471.1 ± 33.6 | 0.000 |
| Three years after surgery | n=24 | n=44 | |
| Residual urine | 36.3 ± 20.7 | 51.3 ± 29.6 | 0.036 |
| Maximum flow rate (mL/sec) | 17.5 ± 3.8 | 15.9 ± 3.7 | 0.040 |
| Maximum reservoir capacity (mL) | 413.6 ± 34.8 | 509.1 ± 35.1 | 0.000 |
| Four years after surgery | n=14 | n=33 | |
| Residual urine | 29.8 ± 21.7 | 55.4 ± 37.6 | 0.018 |
| Maximum flow rate (mL/sec) | 18.6 ± 2.8 | 17.0 ± 3.1 | 0.023 |
| Maximum reservoir capacity (mL) | 443.9 ± 28.8 | 543.2 ± 36.5 | 0.000 |
| Five years after surgery | n=7 | n=21 | |
| Residual urine | 23.1 ± 15.3 | 64.0 ± 58.0 | 0.036 |
| Maximum flow rate (mL/sec) | 19.0 ± 2.3 | 14.2 ± 3.5 | 0.001 |
| Maximum reservoir capacity (mL) | 456.0 ± 53.6 | 581.2 ± 41.3 | 0.037 |

This difference may be due to the inconsistent definition of urinary incontinence and the inconsistent initial bladder capacity.

Other data also show that the control of urination during the day is better than that at night (16). It may be related to the loss of local spinal cord reflex arc, decrease of striated muscle tone and nighttime diuresis. In our study, in the observation group, 100% daytime self-control and 88.2% night self-control were in the 5th and 4th year after surgery. In the observation group, 90.9% of the daytime self-control and 76.4% of the night self-control were in the 3rd year after the operation. Adequate capacity and high compliance of the new bladder may significantly improve the patient's urinary control rate.

In the urodynamic study, the average residual urine volume after urination of the new bladder in observation group 1 gradually decreased with the extension of follow-up time, and was 22.4 ± 19.8 mL at the fifth year after surgery. The average residual urine volume of observation group 2 entered a plateau after a period of decrease, and even tended to rise. The maximum bladder capacity of the two groups of neobladder was gradually increasing, and by the fifth year of follow-up, they were 456.0 ± 53.6 and 581.2 ± 41.3 mL, respectively. The maximum urine flow rate of the two groups of patients gradually increased with the recovery of the external sphincter strength and other factors, and the maximum values were 19.0 ± 2.3 and 17.0 ± 3.1 mL/sec. From the analysis of urodynamic data, the bladder function of observation group 2 gradually recovered after the operation, but by the fourth to five years of follow-up, the increase of residual urine and the decrease of maximum urine flow rate indicated the decline of bladder function. The possible reason is that the new bladder is too large. Although the longest follow-up time in this study was 5 years, there were still some patients with short follow-up time. At the same time, the small number of enrolled patients is also the weakness of this study.

Radical cystectomy and orthotopic neobladder surgery still have obvious complications. And the reporting rate and types of

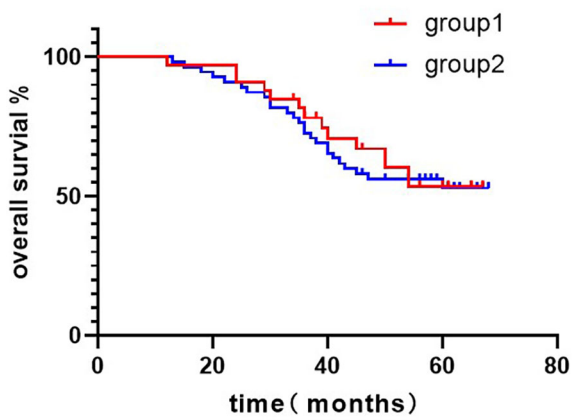


FIGURE 1
Kaplan–Meier curves of overall survival comparison in patients treated with group 1 versus group 2. 5-years-OS = (group 1: 53.60%, group 2: 52.80%; P = 0.657; HR = 0.855; 95CI = 0.429 to 1.705). Log-rank test: P=0.655.

complications vary greatly (17–20). Early complications include bleeding, pulmonary complications, gastrointestinal and urinary system infections. Late complications are mainly affected by urinary diversion, including pyelonephritis, renal atrophy, bladder stones, and anastomotic stenosis.

The incidence of neo-bladder bacterial colonization associated with residual urine is 40-80% (21–23). Then, as the disease progresses, it gradually develops into obvious pyelonephritis (24). In our study, 4 and 5 patients in the two groups had postoperative pyelonephritis (Table 2). In the case of excluding urinary tract obstruction, urinary bacterial culture and drug sensitivity can better control the infection.

Studies have shown that anastomotic stenosis is one of the serious complications of orthotopic neobladder, and its incidence is 2.49% (25, 26). Severe ureterintestinal anastomosis stenosis leads to moderate to severe hydronephrosis and affects renal function. Urethral neobladder anastomotic stenosis will affect the emptying of urine and can be treated by dilatation of the urethra or a second operation. Another complication that affects urine emptying is neurogenic bladder. Due to radical cystectomy to clean the pelvic floor lymph nodes, it may cause damage to the proximal urethra and pelvic floor nerves. Such patients need long-term pelvic floor muscle exercises. One of the main concerns of the stapled reservoirs used in this study is the formation of stone. In Fontana et al (27) and Porena et al (28) study, the median follow-up period of 20 months and stone incidence of 64 months were 6% and 16%. In our study, the incidence of bladder stones was higher than in previous studies. Although titanium staples are conducive to the formation of stones, we have also noticed in this study that some stone patients have urinary tract infections and urinary retention at the same time. Therefore, we believe that there are many reasons for the formation of stones, not only related to titanium staples, but urinary tract infection is also an important factor.

## Conclusion

In short, orthotopic ileal neobladder is technically feasible. Our results show that the 25-35cm ileum-made bladder takes longer to maintain good bladder function than the 35-45cm ileum-made bladder to maintain good bladder function. It provides a new choice for the selection of the truncated length of the orthotopic ileum bladder. Although the follow-up time of observation group 1 was shorter than that of observation group 2, and with the extension of time, the number of people included in observation group 2 gradually decreased, and there was a certain selection deviation in the data, which made the results of the study have certain limitations. However, this study has certain reference significance for obtaining the ileum length from the orthotopic ileal neobladder.

## Data availability statement

The raw data supporting the conclusions of this article will be made available by the authors, without undue reservation.

## Ethics statement

This study was reviewed and approved by Ethics Committee of Zhejiang Provincial People's Hospital (2020QT261). The

patients/participants provided their written informed consent to participate in this study.

## Author contributions

Study conception and design: BZ and ZL. Acquisition of data; JW and HW. Analysis and interpretation of data: BZ and ZL. Drafting of manuscript: BZ and ZL. Critical revision of manuscript: PZ and DZ. All authors contributed to the article and approved the submitted version.

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## Conflict of interest

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

## Publisher's note



## Supplementary material

The Supplementary Material for this article can be found online at: https://www.frontiersin.org/articles/10.3389/fonc.2022.972676/full#supplementary-material

## References

1. Lee RK, Abol-Enein H, Artibani W, Bochner B, Dalbagni G, Daneshmand S, et al. Urinary diversion after radical cystectomy for bladder cancer: Options, patient selection, and outcomes. *Bju Int* (2014) 113(1):11–23. doi: 10.1111/bju.12121

2. Koie T, Hatakeyama S, Yoneyama T, Hashimoto Y, Kamimura N, Ohyama C. Uterus-, fallopian tube-, ovary-, and vagina-sparing cystectomy followed by U-shaped ileal neobladder construction for female bladder cancer patients: oncological and functional outcomes. *Urology* (2010) 75(6):1499–503. doi: 10.1016/j.urology.2009.08.083

3. Koie T, Hatakeyama S, Yoneyama T, Ishimura H, Yamato T, Ohyama C. Experience and functional outcome of modified ileal neobladder in 95 patients. *Int J Urol* (2006) 13(9):1175–9. doi: 10.1111/j.1442-2042.2006.01525.x

4. Abreu SC, Araújo MB, Silveira RA, Regadas RP, Pinheiro DG, Messias FI, et al. Laparoscopic-assisted radical cystectomy with U-shaped orthotopic ileal neobladder constructed using nonabsorbable titanium staples. *Urology* (2006) 68 (1):193–7. doi: 10.1016/j.urology.2006.02.011

5. Witjes JA, Lebret T, Compérat EM, Cowan NC, De Santis M, Bruins HM, et al. Updated 2016 EAU guidelines on muscle-invasive and metastatic bladder cancer. *Eur Urol* (2017) 71(3):462–745. doi: 10.1016/j.eururo.2016.06.020

6. Hautmann RE, Miller K, Steiner U, Wenderoth U. The ileal neobladder: 6 years of experience with more than 200 patients. *J Urol* (1993) 150(1):40–5. doi: 10.1016/s0022-5347(17)35392-2

7. Studer UE, Danuser H, Thalmann GN, Springer JP, Turner WH. Antireflux nipples or afferent tubular segments in 70 patients with ileal low pressure bladder substitutes: Long-term results of a prospective randomized trial. *J Urol* (1996) 156 (6):1913–7. doi: 10.1097/00005392-199612000-00004

8. Mastroianni R, Ferriero M, Tuderti G, Anceschi U, Bove AM, Brassetti A, et al. Open radical cystectomy versus robot-assisted radical cystectomy with intracorporeal urinary diversion: Early outcomes of a single-center randomized controlled trial. *J Urol* (2022) 207(5):982–92. doi: 10.1097/JU.0000000000002422

9. Mastroianni R, Tuderti G, Anceschi U, Bove AM, Brassetti A, Ferriero M, et al. Comparison of patient-reported health-related quality of life between open radical cystectomy and robot-assisted radical cystectomy with intracorporeal urinary diversion: Interim analysis of a randomised controlled trial. *Eur Urol Focus* (2022) 8(2):465–71. doi: 10.1016/j.euf.2021.03.002

10. Dong L, Qin Y, Ya L, Liang C, Tinghui H, Pinlin H, et al. Bayesian Network analysis of open, laparoscopic, and robot-assisted radical cystectomy for bladder cancer. *Med (Baltimore)* (2020) 99(52):e23645. doi: 10.1097/MD.0000000000023645

11. Bai YC, Wang S, Zheng W, Li EH, Quan J, Wei F, et al. Clinical outcome of laparoscopic versus robot-assisted radical cystectomy for patients with bladder cancer: A retrospective study. *BMC Surg* (2021) 21(1):388–3914. doi: 10.1186/s12893-021-01382-1

12. Nam JK, Kim TN, Park SW, Lee SD, Chung MK. The studer orthotopic neobladder: Long-term (More than 10 years) functional outcomes, urodynamic features, and complications. *Yonsei Med J* (2013) 54(3):690–5. doi: 10.3349/ymj.2013.54.3.690

13. Ahmadi H, Skinner EC, Simma-Chianget V, Miranda G, Cai J, Penson DF, et al. Urinary functional outcome following radical cystoprostatectomy and ileal neobladder reconstruction in male patients. *J Urol* (2013) 189(5):1782–8. doi: 10.1016/j.juro.2012.11.078

14. Hautmann RE, Petriconi RD, Gottfried HW, Kleinschmidt K, Mattes R, Paiss T, et al. The ileal neobladder: Complications and functional results in 363 patients after 11 years of followup. *J Urol* (1999) 161(2):422–7. doi: 10.1016/s0022-5347(01)61909-8

15. Gilbert SM, Wood DP, Dunn RL, Weizer AZ, Lee CT, Montie JE, et al. Measuring health-related quality of life outcomes in bladder cancer patients using the bladder cancer index (BCI). *Cancer-Am Cancer Soc* (2007) 109(9):1756–62. doi: 10.1002/cncr.22556

16. Canda AE, Atmaca AF, Altinova S, Akbulut Z, Balbay MD. Robot-assisted nerve-sparing radical cystectomy with bilateral extended pelvic lymph node dissection (PLND) and intracorporeal urinary diversion for bladder cancer: Initial experience in 27 cases. *Bju Int* (2012) 110(3):434–44. doi: 10.1111/j.1464-410X.2011.10794.x

17. Studer UE, Burkhard FC, Schumacher M, Kessler TM, Thoeny H, Fleischmann A, et al. Twenty years experience with an ileal orthotopic low pressure bladder substitute–lessons to be learned. *J Urol* (2006) 176(1):161–6. doi: 10.1016/S0022-5347(06)00573-8

18. Lawrentschuk N, Colombo R, Hakenberg OW, Lerner SP, Månsson W, Sagalowsky A, et al. Prevention and management of complications following radical cystectomy for bladder cancer. *Eur Urol* (2010) 57(6):983–1001. doi: 10.1016/j.eururo.2010.02.024

19. Novotny V, Hakenberg OW, Wiessner D, Heberling U, Litz RJ, Oehlschlaeger S, et al. Perioperative complications of radical cystectomy in a

contemporary series. *Eur Urol* (2007) 51(2):397–401. doi: 10.1016/j.eururo.2006.06.014

20. Parekh DJ, Gilbert WB, Koch MO, Smith JA. Continent urinary reconstruction versus ileal conduit: A contemporary single-institution comparison of perioperative morbidity and mortality. *Urology* (2000) 55(6):852–5. doi: 10.1016/s0090-4295(99)00619-6

21. Wullt B, Holst E, Stevenet K, Carstensen J, Pedersen J, Gustafsson E, et al. Microbial flora in ileal and colonic neobladders. *Eur Urol* (2004) 45(2):233–9. doi: 10.1016/j.eururo.2003.09.002

22. Wood DP, Bianco FJ, Pontes JE, Heath MA, DaJusta D. Incidence and significance of positive urine cultures in patients with an orthotopic neobladder. *J Urol* (2003) 169(6):2196–9. doi: 10.1097/01.ju.0000067909.98836.91

23. Akerlund S, Campanello M, Kaijser B, Jonsson O. Bacteriuria in patients with a continent ileal reservoir for urinary diversion does not regularly require antibiotic treatment. *Br J Urol* (1994) 74(2):177–81. doi: 10.1111/j.1464-410x.1994.tb16582.x

24. Molander U, Sundh V, SteenUrinary B. Urinary incontinence and related symptoms in older men and women studied longitudinally between 70 and 97 years of age. A population study. *Arch Gerontol Geriat* (2002) 35(3):237–44. doi: 10.1016/s0167-4943(02)00032-8

25. Shaaban AA, Mosbah A, El-Bahnasawy MS, Madbouly K, Ghoneim MA. The urethral kock pouch: Long-term functional and oncological results in men. *Bju Int* (2003) 92(4):429–35. doi: 10.1046/j.1464-410x.2003.04346.x

26. Jensen JB, Lundbeck F, Jensen KM. Complications and neobladder function of the hautmann orthotopic ileal neobladder. *Bju Int* (2006) 98(6):1289–94. doi: 10.1111/j.1464-410X.2006.06449.x

27. Fontana D, Bellina M, Fasolis G, Frea B, Scarpa RM, Mari M, et al. Y-neobladder: An easy, fast, and reliable procedure. *Urology* (2004) 63(4):699–703. doi: 10.1016/j.urology.2003.11.015

28. Porena M, Mearini L, Zucchi A, Zingaro MD, Mearini E, Giannantoni A. Perugia ileal neobladder: Functional results and complications. *World J Urol* (2012) 30(6):747–52. doi: 10.1007/s00345-012-0985-z

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# Prognostic value and immunological role of BAIAP2L2 in liver hepatocellular carcinoma: A pan-cancer analysis

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**Background:** In recent years, the role of BAI1-associated protein 2-like 2 (BAIAP2L2) in the prognosis and immune microenvironment of various cancers has attracted increasing attention. However, its clinical value and immune infiltration in liver hepatocellular carcinoma (LIHC) remain unclear.
**Objective:** To investigate the prognostic value of BAIAP2L2 and its correlation with immune infiltration in LIHC, we conducted corresponding data mining.
**Methods:** In this study, The Cancer Genome Atlas, GTEx, StarBase, UALCAN, TIMER, GEPIA, Human Protein Atlas, Kaplan–Meier Plotter, cBioPortal, LinkedOmics, STRING and BioGPS databases were used to analyze BAIAP2L2 in cancers. Logistic regression and Cox regression were performed to analyze the correlation between clinical features and BAIAP2L2 expression in LIHC. In addition, the diagnostic and prognostic values of BAIAP2L2 in LIHC were determined by receiver operating characteristic (ROC) curves and nomograms. Single-sample gene set enrichment analysis (ssGSEA), BioGPS and TIMER were used to analyze the correlation between BAIAP2L2 and immune infiltration. More importantly, quantitative real-time polymerase chain reaction was used to verify BAIAP2L2 expression in a liver cancer cell

Abbreviations
ACC, adrenocortical carcinoma; AFP, alpha fetoprotein; AUC, area under the curve; BAIAP2L2, BAI1-associated protein 2-like 2; BLCA, bladder urothelial carcinoma; BPs, biological processes; BRCA, breast invasive carcinoma; CCs, cellular components; CESC, endocervical adenocarcinoma; CHOL, cholangiocarcinoma; CI, confidence intervals; CNA, copy number alteration; DCs, dendritic cells; DFS, disease-free survival; DLBC, diffuse large B-cell lymphoma; EGA, European Genome-Phenome Archive; ESCA, esophageal carcinoma; FBS, fetal bovine serum; GEO, Gene Expression Omnibus; GO, Gene Ontology; GSEA, Gene Set Enrichment Analysis; HNSC, head and neck squamous cell carcinoma; HR, hazard ratio; I-BAR, Inverse Bin-Amphiphysin-Rvs; KEGG, Kyoto Encyclopedia of Genes and Genomes; KIRC, kidney renal clear cell carcinoma; KIRP, kidney renal papillary cell carcinoma; LAML, acute myeloid leukemia; LGG, lower grade glioma; LIHC, liver hepatocellular carcinoma; LUAD, lung adenocarcinoma; LUSC, lung squamous cell carcinoma; MEM, Minimum Essential Medium; MESO, mesothelioma; MFs, molecular functions; NK, natural killer; OS, overall survival; OV, ovarian serous cystadenocarcinoma; PAAD, pancreatic adenocarcinoma; PCPG, pheochromocytoma and paraganglioma; PPI, protein–protein interaction; PRAD, prostate adenocarcinoma; ROC, receiver operating characteristic; SKCM, skin cutaneous melanoma; ssGSEA, single-sample gene set enrichment analysis; STAD, stomach adenocarcinoma; TAMs, tumor-associated macrophages; TCGA, The Cancer Genome Atlas; Tfh, T follicular helper; TGCTs, testicular germ cell tumors; THYM, thymoma; TME, tumor microenvironment; UCEC, uterine corpus endometrial carcinoma; UCS, uterine carcinosarcoma; UVM, uveal melanoma

line and a normal cell line. Visualization of data was mostly achieved using R language, version 3.6.3.

**Results:** High BAIAP2L2 levels indicated poor overall survival (OS) and disease-free survival (DFS) of patients with LIHC. Abnormally increased expression of BAIAP2L2 in LIHC may be the result of both genetic alterations and lower DNA methylation levels. Furthermore, Cox regression analysis showed that high BAIAP2L2 expression was an independent risk factor for OS and DFS in patients with liver cancer. ROC curves and nomograms also confirmed the diagnostic and prognostic values of BAIAP2L2 in LIHC. Additionally, a PPI network of BAIAP2L2 was established and results implyed that BAIAP2L2 interacts with MTSS1, AMPH, FCHO1, SYT9, PDK2, MTSS1L, PM20D1, CHST4 and PALM3. ssGSEA showed that BAIAP2L2 was associated with T cells and natural killer cells. Simultaneously, the TIMER database showed that the expression of BAIAP2L2 in LIHC was positively correlated with tumor infiltrating cells, including B cells, CD8+ T cells, CD4+ T cells, macrophages, neutrophils and dendritic cells.

**Conclusions:** Through pan-cancer analysis, prognostic and immunological value of BAIAP2L2 in LIHC was identified. This is the first report on the potential of BAIAP2L2 as a prognostic biomarker and its correlation with immune infiltration in LIHC.

KEYWORDS

BAIAP2L2, liver hepatocellular carcinoma (LIHC), prognostic value, immune infiltration, pan-cancer analysis

## Introduction

Due to the increasing prevalence of established risk factors, such as the growth and aging of the population, smoking, being overweight, lacking physical activity, urbanization and changes in reproductive patterns resulting from economic development, the incidence of cancer is gradually rising and has become a huge burden on society (1). Liver hepatocellular carcinoma (LIHC; also known as HCC) is the sixth most commonly diagnosed cancer and the fourth leading cause of cancer mortality in the world, with approximately 841,000 new cases and 782,000 deaths annually (2). In the past 30 years, the incidence of HCC has been on the rise worldwide. The World Health Organization estimates that more than 1 million patients will die from liver cancer in 2030 (3). The highest incidence rates of HCC were mainly observed in Northern and Western Africa (Egypt, the Gambia, Guinea) and Eastern and Southeastern Asia (Mongolia, Cambodia, and Vietnam) (2). The dominant risk factors for HCC are chronic infection with hepatitis B virus or hepatitis C virus, nonalcoholic fatty liver disease, alcohol consumption, cigarette smoking and environmental toxins (4). The treatment of HCC mainly includes surgical resection, liver transplantation, vascular intervention and radiofrequency ablation (5). Despite the emergence of targeted therapy, advanced-stage LIHC remains largely incurable due to low response rate and therapeutic resistance (6). Moreover, most of tumours response to immunotherapies is either non-existent or short-lived (7). To improve the survival rate of LIHC patients, new therapeutic targets will still need to be discovered so that existing drug options can be increased and a better understanding of the underlying mechanisms leading to drug resistance can be gained.

BAIAP2L2 (BAI1-associated protein 2-like 2; also known as Pinkbar) is located on chromosome 22q131 (8). Along with BAIAP2L1, IRSp53, MTSS1 and MTSS1L, BAIAP2L2 belongs to the I-BAR (Inverse Bin-Amphiphysin-Rvs) subfamily (9, 10). They have different isoforms, but all contain an N-terminal I-BAR domain. In recent years, the I-BAR family has been found to be related to the occurrence of tumors (11, 12). Overexpression of IRTKs was negatively correlated with overall survival in patients with gastric cancer (13). BAIAP2L1 is also a potential biomarker in ovarian cancer (14) and IRSP53 plays an important role in regulating the motility/invasion of cancer cells (15, 16). Studies have shown that MTSS1 is notably downregulated during the progression of gastric cancer (17), and hypermethylated MTSS1 can promote the migration of prostate cancer (18). The key role of I-BAR family members in the carcinogenesis process. Although BAIAP2L2 was associated with the development of various cancers, including osteosarcoma (19), gastric cancer (20), Prostate Cancer (21) and lung cancer (22). However, no studies have reported the relationship between BAIAP2L2 and liver cancer.

In this study, we comprehensively analyzed the expression profile and prognostic value of BAIAP2L2 in 33 types of cancer, and found that BAIAP2L2 was highly expressed in LIHC. Overexpression of BAIAP2L2 is associated with poor prognosis of LIHC. Results also illustrated the immunological role of BAIAP2L2 in LIHC.

# Materials and methods

## Gene expression analysis

The "Diff Exp" module in the TIMER database (https://cistrome.shinyapps.io/timer/) allows us to study the differential expression of BAIAP2L2 in The Cancer Genome Atlas (TCGA) between tumor and adjacent normal tissues (23, 24). Distributions of gene expression levels are displayed using box plots, with statistical significance of differential expression evaluated using the Wilcoxon test. The GTEx database contains data for normal tissue. If the normal sample size of TCGA is insufficient, we will combine GTEx and TCGA to analyze the differential expression of BAIAP2L2 in tumors. Additionally, StarBase (http://starbase.sysu.edu.cn/) (25), a comprehensive online tool, was also applied to analyze gene expression. The "ENCORI Pan-Cancer Analysis Platform" in the StarBase database is designed to analyze the gene expression profile of 32 cancer types. The expression data of cancers were downloaded from TCGA project *via* Genomic Data Commons Data Portal. Then, the Venn diagrams of data from three database were plotted using the "ggplot2" R package. Finally, the UALCAN database (http://ualcan.path.uab.edu/analysis.html) [26] and Human Protein Atlas (HPA) databases (https://www.proteinatlas.org/) (27) were used to verify the differentiation of BAIAP2L2 expression levels between tumors and normal tissues. Adjusted $p < 0.05$ and |log2fold change (FC)| > 1 were chosen as the cutoff criteria.

## Survival analysis

To explore the prognosis of BAIAP2L2 across cancers, we used the Kaplan–Meier Plotter database (https://kmplot.com/analysis/) (28) and LinkedOmics (http://www.linkedomics.org/) (29) to analyze the effect of BAIAP2L2 on the survival of various cancers. The data sources for the Kaplan–Meier Plotter database include not only the Gene Expression Omnibus (GEO) but also the European Genome-Phenome Archive (EGA) and TCGA. To analyze the prognostic value of a gene, the patient samples are split into two groups according to various quantile expressions. Then, the two patient cohorts are compared by a survival plot, and the hazard ratio with 95% confidence intervals (CI) and logrank *p*-value are calculated. 95% CIs and a $p$ value <0.05 were considered statistically significant.

## CNA and DNA methylation alteration analysis

The cBioPortal for cancer genomics (https://www.cbioportal.org/) was used to query the BAIAP2L2 characteristics of genetic mutations (30, 31). The copy number alteration (CNA) data and mutation type were displayed in the "Cancer Types Summary" module of TCGA database. DNA methylation levels of the BAIAP2L2 promoter in normal and tumor tissues were analyzed using the UALCAN database (http://ualcan.path.uab.edu/index.html). TCGA-assembler pipeline was used to download TCGA DNA methylation data generated using the Illumina HumanMethylation450 BeadChip. Downloaded data were further processed to calculate an average methylation (beta) value for each gene, considering CpG sites located in the promoter region of the gene *via* the TCGA-assembler (32).

## BAIAP2L2 expression-correlated gene and protein analysis

We predicted the genes and proteins interacting with BAIAP2L2 using LinkedOmics and the STRING database (https://string-db.org/) (33), respectively. To further explore the biological functions of BAIAP2L2 in LIHC, Gene Set Enrichment Analysis (GSEA) was used to analyze the Gene Ontology (GO) terms and Kyoto Encyclopedia of Genes and Genomes (KEGG) pathways. GO analysis is a powerful bioinformatics tool used to identify biological processes (BPs), cellular components (CCs) and molecular functions (MFs). The main goal of the KEGG database project is to assign functional meaning to genes and genomes at the molecular and higher levels. A protein–protein interaction (PPI) network of BAIAP2L2 was generated using the STRING database.

## Immune infiltration analysis

Immune infiltration analysis of LIHC was performed using single-sample gene set enrichment analysis (ssGSEA) in the "GSVA" R package, and the infiltration levels of 24 types of immune cells were quantified from gene expression profiles. The TIMER database and GEPIA databases (http://gepia.cancer-pku.cn/) (34) were used to explore the correlation between BAIAP2L2 expression and immune infiltration. We utilized the "Gene" module to estimate the correlation between BAIAP2L2 expression and immune infiltration levels (B cells, CD4+ T cells, CD8+ T cells, neutrophils, macrophages and dendritic cells). Then, the "Correlation" module was applied to analyze the association between BAIAP2L2 and prognosis-related immune cell markers to further estimate the potential infiltrating immune cell subtypes. The correlation coefficient was determined by the Spearman method, and *p values* were corrected using the Benjamini-Hochberg method. Furthermore, BioGPS (http://biogps.org/) is a centralized gene annotation portal that enables researchers to access distributed gene annotation

resources (35). We used this database to display the level of BAIAP2L2 mRNA expression in human tissues and immune cells.

## Cell culture and quantitative real-time polymerase chain reaction (qRT–PCR) of cell lines

The human hepatocarcinoma cell line HepG2 and human normal liver cell line LO2 were cultured in Minimum Essential Medium (MEM, Procell) and Roswell Park Memorial Institute 1640 (RPMI 1640, Procell), respectively, containing 10% fetal bovine serum (FBS, Excell Bio) and antibiotics (100 units/ml penicillin and 100 μg/ml streptomycin) at 37°C and 5% CO2 in an incubator. qRT–PCR was conducted to evaluate gene expression. Total RNA was extracted from cell lines with TRIzol reagent in accordance with the manufacturer's instructions. Using a reverse transcription kit, the RNA was reverse transcribed into cDNA, and qRT–PCR analyses were quantified with SYBR Green (VAZYME). BAIAP2L2 expression was calculated based on the 2−ΔΔCt method with actin as an internal reference. qRT–PCR was performed in triplicate using samples derived from three independent experiments. Primers for BAIAP2L2 (forward: 5′-AGTTCATCAAAGACAGCCGC-3′, reverse: 5′-CAGGTGCTTCTCTGCTAGGA-3′) and *β*-actin (forward: 5′-CACGATGGAGGGGCCGGACTCATC-3′, reverse: 5′-TAAAGACCTCTATGCCAACACAGT-3′) were used for qRT–PCR.

## Statistical analysis

Most of the statistical analyses were performed using the bioinformatic tools mentioned above. The results were shown as the mean ± SD. IBM SPSS statistics 26.0 software was utilized for statistical analysis. A $p$ value <0.05 was considered statistically significant.

# Results

## BAIAP2L2 is universally over-expressed in human pan-cancer

BAIAP2L2 has been proved to be abnormally expressed in cancers (36, 37). We used TIMER and StarBase database to demonstrate BAIAP2L2 expression in 33 types of human cancer. Data from the TIMER database showed that BAIAP2L2 expression was significantly increased in 10 types of cancer, including bladder urothelial carcinoma (BLCA), cholangiocarcinoma (CHOL), esophageal carcinoma (ESCA), head and neck squamous cell carcinoma (HNSC), kidney renal clear cell carcinoma (KIRC), LIHC, lung adenocarcinoma (LUAD), lung squamous cell carcinoma (LUSC), prostate adenocarcinoma (PRAD) and stomach adenocarcinoma (STAD) (**Figure 1A**, $p < 0.05$). Because some cancers in the TIMER database did not have normal tissue, we used the GTEX database combined with the TCGA database to supplement these data. The results showed that BAIAP2L2 was obviously increased in some cancers, including cervical squamous cell carcinoma and endocervical adenocarcinoma (CESC), lymphoid neoplasm diffuse large B-cell lymphoma (DLBC), acute myeloid leukemia (LAML), ovarian serous cystadenocarcinoma (OV), skin cutaneous melanoma (SKCM), pancreatic adenocarcinoma (PAAD), thymoma (THYM) and uterine carcinosarcoma (UCS) (**Figure 1B**, $p < 0.05$).

From StarBase database, we discovered that BAIAP2L2 was evidently upregulated in 14 types of cancer, including BLCA, breast invasive carcinoma (BRCA), CHOL, ESCA, HNSC, KIRC, kidney renal papillary cell carcinoma (KIRP), LIHC, LUAD, LUSC, PAAD, PRAD, STAD and uterine corpus endometrial carcinoma (UCEC) (**Figure 1C**, $p < 0.05$). By comparing results from the three databases (TIMER, GTEX and STARBASE), we concluded that BAIAP2L2 was generally overexpressed across 11 types of human cancer, including BLCA, CHOL, ESCA, HNSC, KIRC, LIHC, LUAD, LUSC, PAAD, PRAD and STAD (**Figure 4A**).

## BAIAP2L2 expression is closely associated with patient survival in human pan-cancer

Considering that BAIAP2L2 is dysregulated in a variety of cancers, we wanted to know whether its expression is related to the survival of cancer patients. Two databases, the Kaplan–Meier Plotter database and LinkedOmics, were utilized to analyze the relationship between BAIAP2L2 expression and patient overall survival (OS) in 33 types of cancer. The results from the Kaplan–Meier Plotter database demonstrated that a high level of BAIAP2L2 indicated unfavorable OS in CESC ($p = 0.042$), LIHC ($p = 0.0026$), and LUAD ($p = 0.0059$) and good OS in ESCA ($p = 0.018$), KIRC ($p = 0.0036$), and KIRP ($p = 0.032$) (**Figure 2B**). From LinkedOmics, we found that elevated BAIAP2L2 expression predicted worse OS in adrenocortical carcinoma (ACC) ($p = 6.724e-03$), LIHC ($p = 1.108e-03$), LUAD ($p = 1.430e-03$), mesothelioma (MESO) ($p = 3.436e-03$), PRAD ($p = 5.183e-03$), uveal melanoma (UVM) ($p = 1.706e-03$) and good OS in glioma ($p = 2.587e-14$) and brain lower grade glioma (LGG) ($p = 2.797e-04$) (**Figure 2C**).

Next, the Kaplan–Meier Plotter database was used to explore the relationship between BAIAP2L2 levels and patient

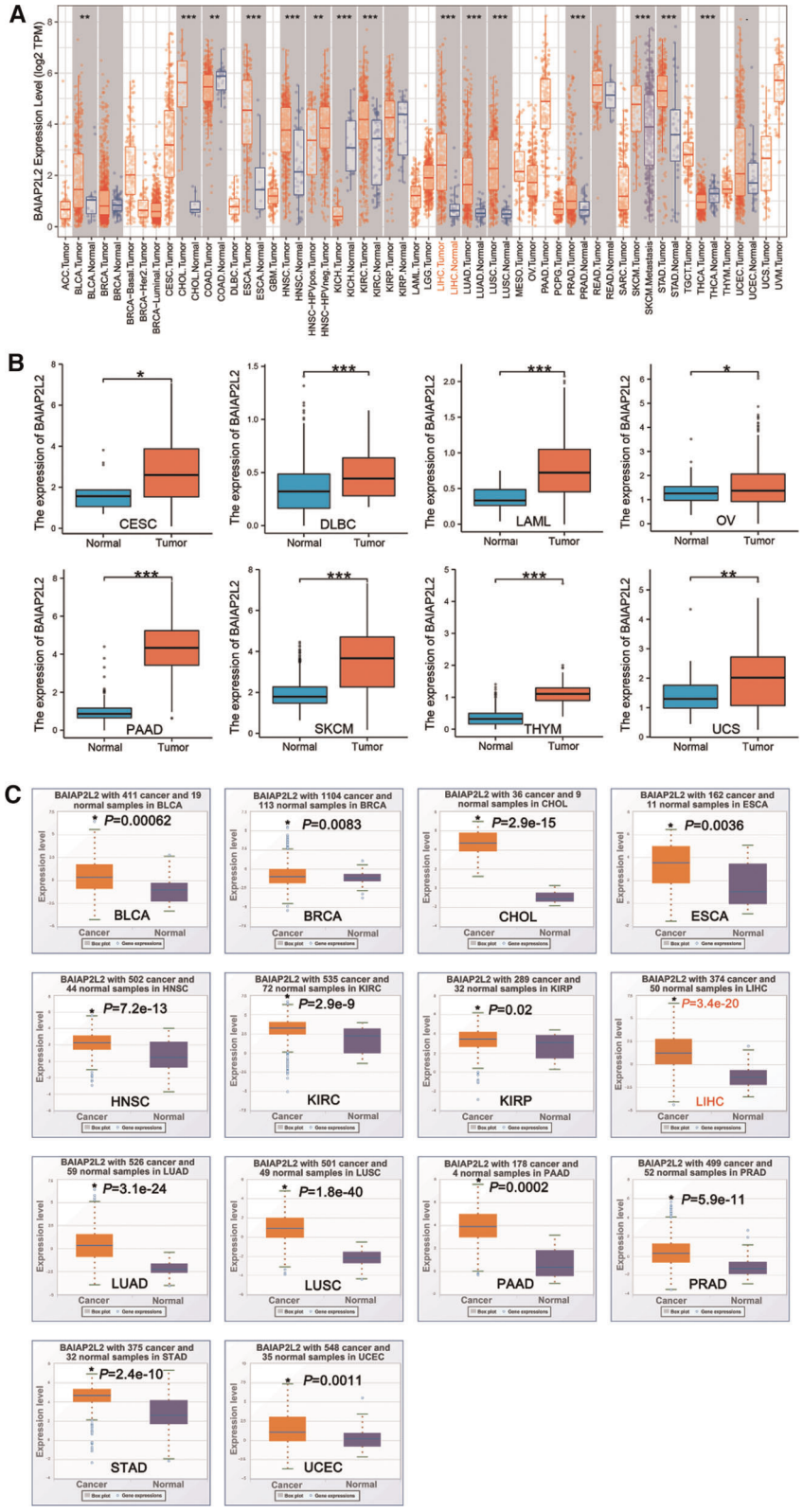


FIGURE 1
BAIAP2L2 expression levels in different types of cancer. (A) BAIAP2L2 expression levels in different types of cancer from TCGA datasets in TIMER. (B) BAIAP2L2 expression levels in different types of cancer from GTEX database combined with the TCGA database. *$p < 0.05$, **$p < 0.01$, ***$p < 0.001$. (C) Upregulated transcriptional level of BAIAP2L2 in pan-cancer samples from the StarBase database. The orange and purple boxes represent cancer and Normal samples, respectively, *$p < 0.05$ compared to the Normal tissue.

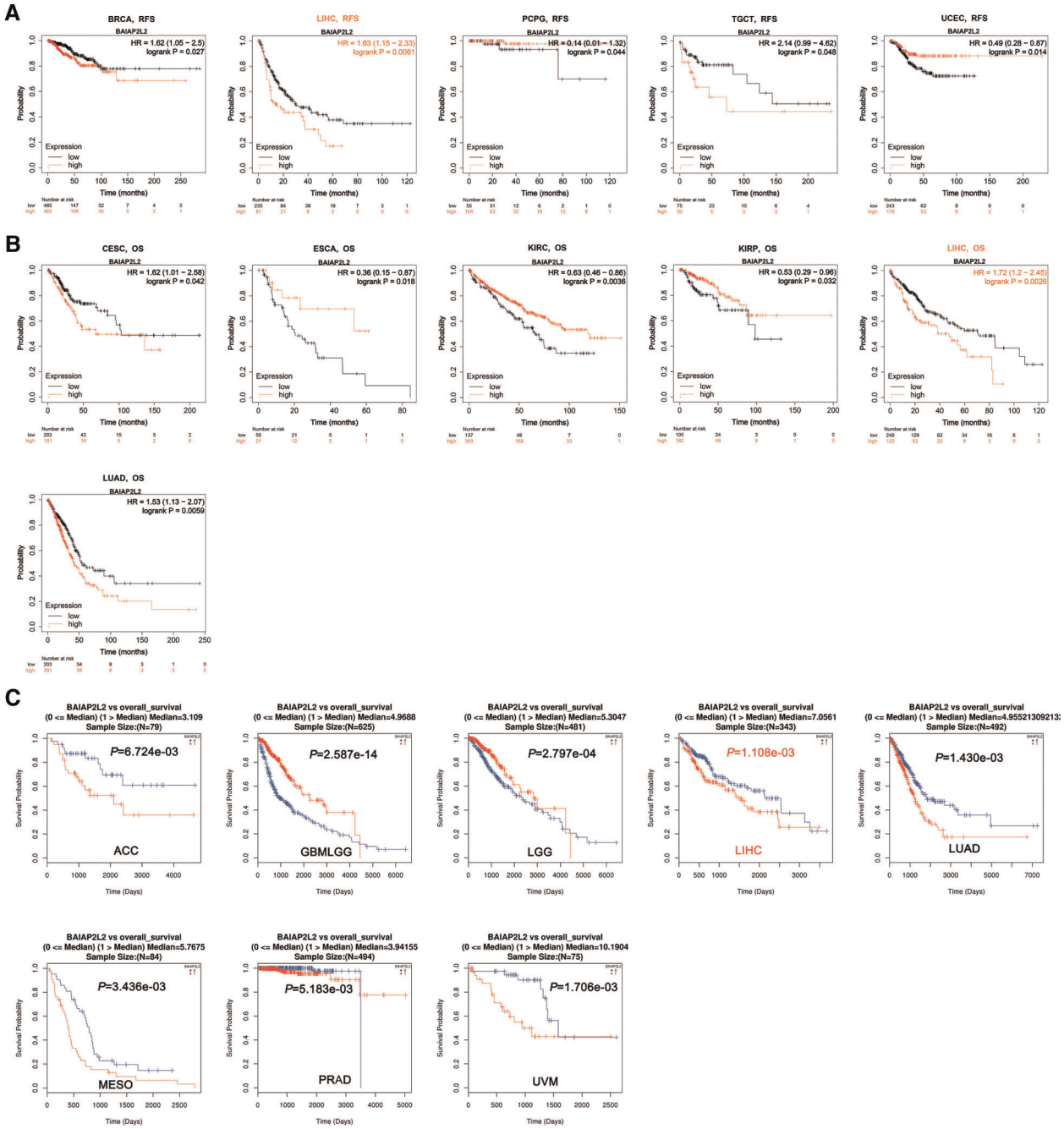


FIGURE 2
Survival analyses of BAIAP2L2 expression in different types of cancer. (A) Survival analysis of RFS in Kaplan–Meier plotter. (B) The survival analysis of OS in Kaplan–Meier plotter. (C) The survival analysis of OS From LinkedOmics.

disease-free survival (DFS) in 33 types of cancer. The results showed that a high level of BAIAP2L2 indicated poor DFS in BRCA ($p = 0.027$), LIHC ($p = 0.0061$), and testicular germ cell tumors (TGCTs) ($p = 0.048$) and good DFS in pheochromocytoma and paraganglioma (PCPG) ($p = 0.044$) and UCEC ($p = 0.014$) (Figure 2A). By comparing the results of Figures 1, 2, we found that high BAIAP2L2 levels were significantly correlated with poor patient OS and DFS in LIHC (Figures 4B,C). These data imply that BAIAP2L2 has potential prognostic value in LIHC.

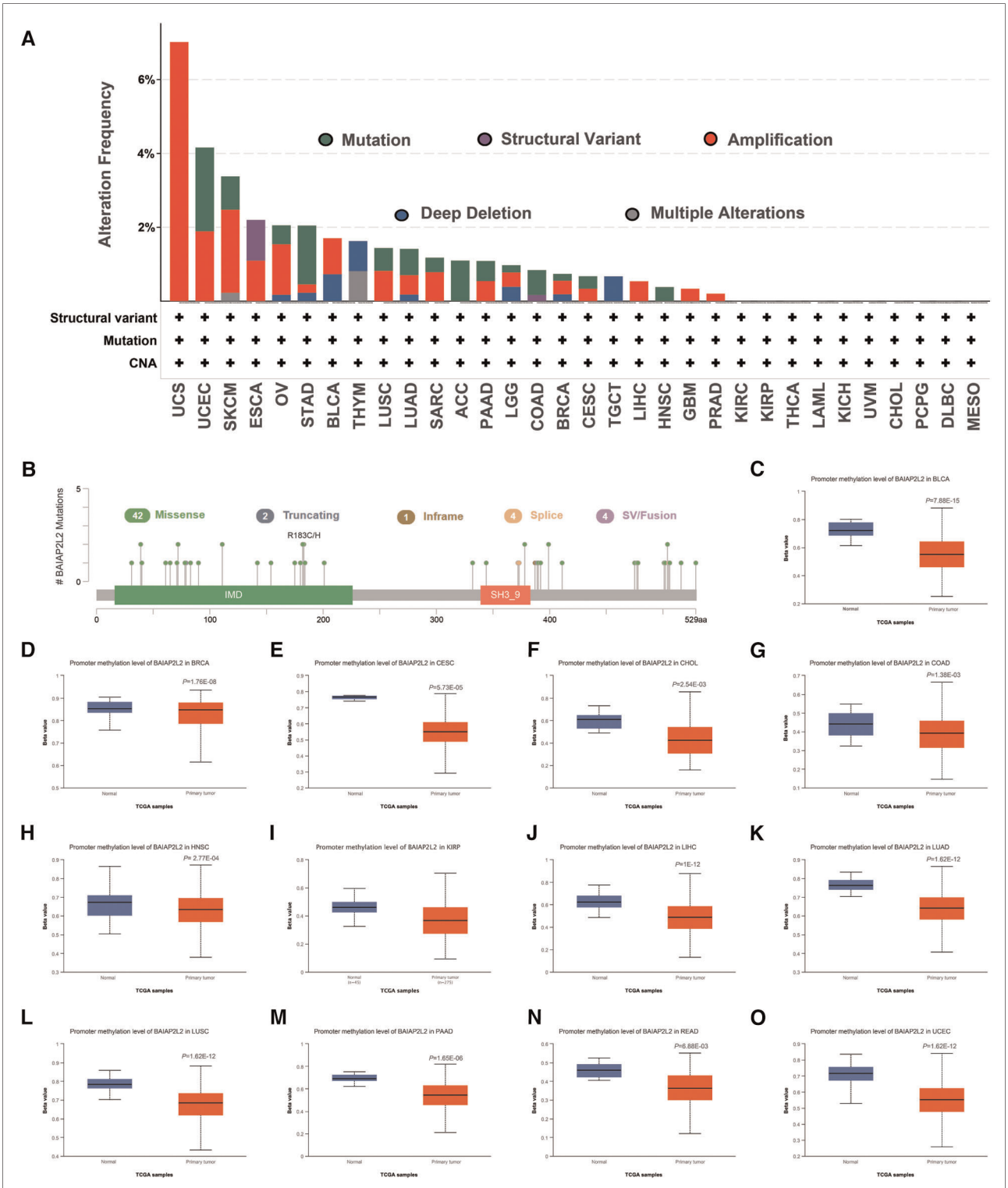


FIGURE 3
BAIAP2L2 mutated landscapes and methylation levels. (**A**) BAIAP2L2 mutation frequency in multiple TCGA pancancer studies according to the cBioPortal database. (**B**) Mutation diagram of BAIAP2L2 in different cancer types across protein domains. (**C–O**) BAIAP2L2 methylation levels were determined by UALCAN, and different beta value cutoffs have been considered to indicate hypermethylation [beta value: 0.7–0.5] or hypomethylation [beta value: 0.3–0.25].

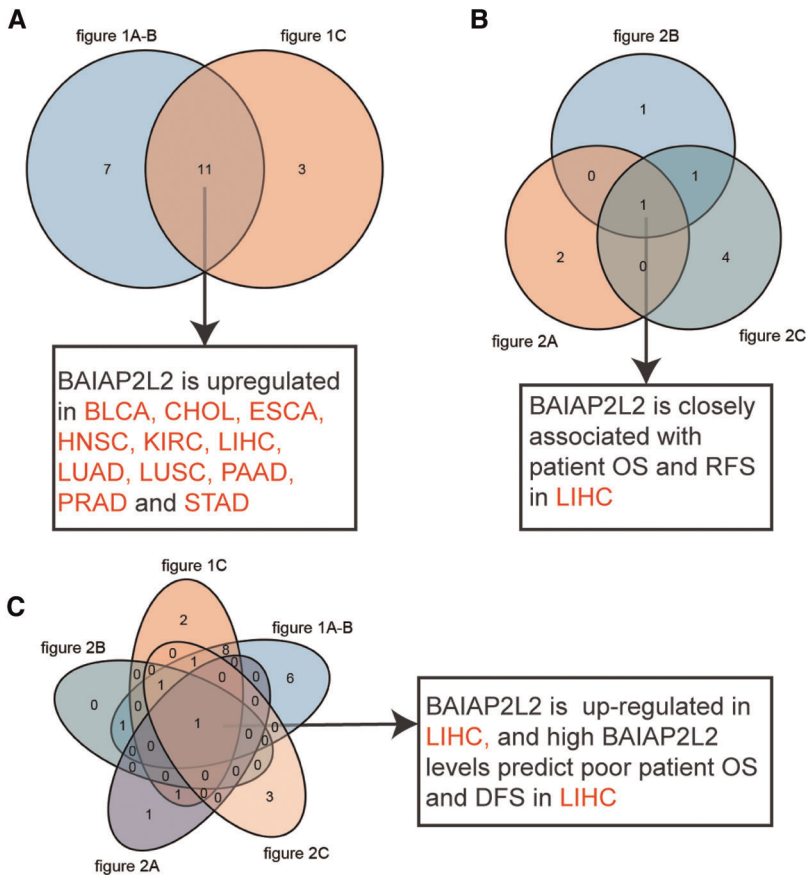


FIGURE 4
Data comparison is shown as Venn diagrams. (**A**) Comparative analysis of data in Figure 1, BAIAP2L2 is overexpressed in BLCA, CHOL, ESCA, HNSC, KIRC, LIHC, LUAD, LUSC, PAAD, PRAD and STAD. (**B**) BAIAP2L2 is closely associated with patient OS and RFS in LIHC (Figure 2). (**C**) High BAIAP2L2 levels predict poor patient OS and DFS in LIHC (Figures 1, 2).

## CNA and DNA methylation alterations of BAIAP2L2 across different human cancers

Genetic and epigenetic changes play a significant role in regulating cancer development and immune tolerance (38). As shown in **Figure 3A**, we can see that elevated BAIAP2L2 expression was accompanied by gene alterations in BLCA, ESCA, LIHC, LUAD, LUSC, HNSC, PAAD, PRAD and STAD. It is worth noting that all LIHC cases with genetic alterations had amplification of BAIAP2L2. The types, sites and case number of the BAIAP2L2 genetic alteration are further presented in **Figure 3B**, and we found that missense mutation of BAIAP2L2 was the primary type of genetic alteration.

DNA methylation is an epigenetic mechanism that can affect the progression of tumors (39). To correlate promoter DNA methylation levels with BAIAP2L2 expression, we explored the differential promoter DNA methylation status of BAIAP2L2 between tumors and adjacent normal tissues by using UALCAN (**Figures 3C–O**). we found that BAIAP2L2 had lower DNA methylation levels in LIHC. Overall, abnormally increased expression of BAIAP2L2 mRNA in LIHC may be the result of both genetic alterations and lower DNA methylation levels.

## BAIAP2L2 is up-regulated and is associated with poor prognosis in LIHC

By comprehensive analysis of the expression (**Figure 1**) and prognosis (**Figure 2**) of BAIAP2L2 across cancers, we found that BAIAP2L2 was overexpressed in LIHC and was closely associated with poor prognosis of liver cancer patients (**Figure 4**), implying its importance in predicting the clinical outcome of LIHC. Therefore, we focused on investigating the function of BAIAP2L2 in LIHC.

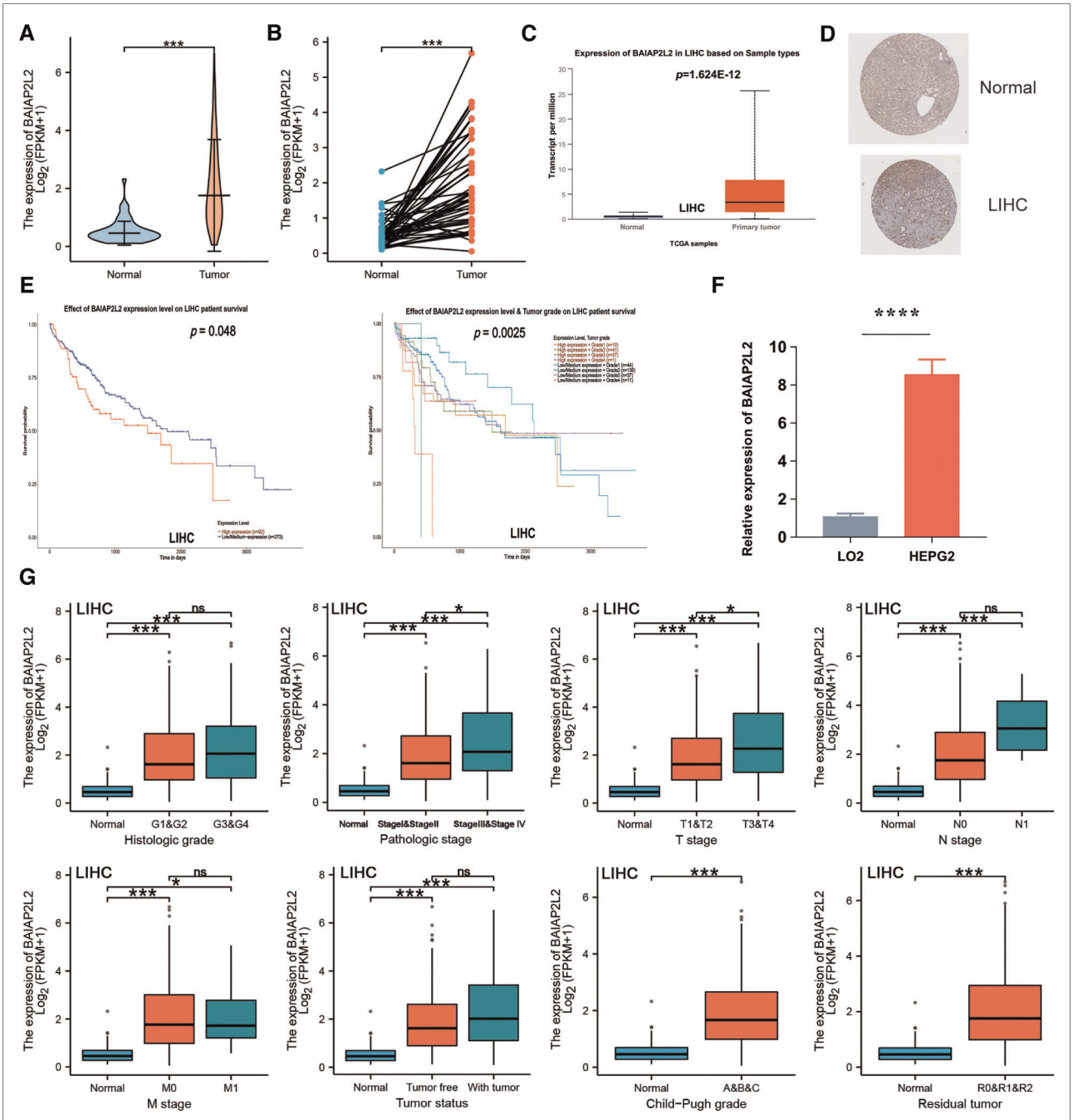


FIGURE 5
Validation of the expression level of BAIAP2L2 and high BAIAP2L2 expression is correlated with clinical characteristics in patients with LIHC. (**A**) The differential expression of BAIAP2L2 in LIHC samples and unpaired adjacent samples from TCGA. (**B**) The differential expression of BAIAP2L2 in 50 LIHC samples and matched adjacent samples from TCGA. (**C**) Validation of BAIAP2L2 at the mRNA level using the UALCAN database. (**D**) Validation of BAIAP2L2 at the protein level using the HPA database. (**E**) Survival analyses of BAIAP2L2 expression in LIHC. (**F**) Validation of BAIAP2L2 at the mRNA level *in vitro* using qRT−PCR analysis. Relative BAIAP2L2 mRNA levels in a LIHC cell line (HEPG2) and a human Normal liver cell line (LO2). (**G**) Box plot assessing BAIAP2L2 expression in patients with LIHC according to different clinical characteristics using the UALCAN database. *$p < 0.05$, **$p < 0.01$, ***$p < 0.001$, ****$p < 0.0001$.

Firstly, BAIAP2L2 expression in LIHC samples and adjacent normal tissues was analyzed through TCGA. BAIAP2L2 expression was observably elevated in LIHC tissues (Figures 5A,B). Then, the results from the UALCAN database showed that BAIAP2L2 was dramatically upregulated in LIHC compared to normal tissues (Figure 5C), and a high level of BAIAP2L2 indicated unfavorable survival probability in LIHC (Figure 5E). In addition, we examined the protein level of

TABLE 1 Correlation between BAIAP2L2 expression and clinicopathologic characteristics of patients with LIHC.

| Characteristic | Low expression of BAIAP2L2 | High expression of BAIAP2L2 | *p* |
|---|---|---|---|
| *n* | 185 | 186 | |
| Gender, *n* (%) | | | 0.001 |
| Female | 45 (12.1%) | 76 (20.5%) | |
| Male | 140 (37.7%) | 110 (29.6%) | |
| T stage, *n* (%) | | | 0.288 |
| T1 | 98 (26.6%) | 83 (22.6%) | |
| T2 | 46 (12.5%) | 48 (13%) | |
| T3 | 34 (9.2%) | 46 (12.5%) | |
| T4 | 5 (1.4%) | 8 (2.2%) | |
| N stage, *n* (%) | | | 0.622 |
| N0 | 127 (49.6%) | 125 (48.8%) | |
| N1 | 1 (0.4%) | 3 (1.2%) | |
| M stage, *n* (%) | | | 1.000 |
| M0 | 131 (48.5%) | 135 (50%) | |
| M1 | 2 (0.7%) | 2 (0.7%) | |
| Pathologic stage, *n* (%) | | | 0.269 |
| Stage I | 92 (26.5%) | 79 (22.8%) | |
| Stage II | 43 (12.4%) | 43 (12.4%) | |
| Stage III | 35 (10.1%) | 50 (14.4%) | |
| Stage IV | 3 (0.9%) | 2 (0.6%) | |
| Histologic grade, *n* (%) | | | 0.016 |
| G1 | 35 (9.6%) | 20 (5.5%) | |
| G2 | 94 (25.7%) | 83 (22.7%) | |
| G3 | 48 (13.1%) | 74 (20.2%) | |
| G4 | 6 (1.6%) | 6 (1.6%) | |
| Residual tumor, *n* (%) | | | 0.320 |
| R0 | 164 (48%) | 160 (46.8%) | |
| R1 | 6 (1.8%) | 11 (3.2%) | |
| R2 | 1 (0.3%) | 0 (0%) | |
| Child–Pugh grade, *n* (%) | | | 0.427 |
| A | 114 (47.7%) | 103 (43.1%) | |
| B | 13 (5.4%) | 8 (3.3%) | |
| C | 0 (0%) | 1 (0.4%) | |
| Adjacent hepatic tissue inflammation, *n* (%) | | | 0.486 |
| None | 56 (23.9%) | 61 (26.1%) | |
| Mild | 53 (22.6%) | 46 (19.7%) | |
| Severe | 11 (4.7%) | 7 (3%) | |
| Vascular invasion, *n* (%) | | | 0.104 |
| No | 112 (35.6%) | 94 (29.8%) | |
| Yes | 48 (15.2%) | 61 (19.4%) | |
| Fibrosis Ishak score, *n* (%) | | | 0.395 |
| 0 | 41 (19.3%) | 33 (15.6%) | |
| ½ | 21 (9.9%) | 10 (4.7%) | |
| ¾ | 13 (6.1%) | 15 (7.1%) | |
| 5/6 | 42 (19.8%) | 37 (17.5%) | |
| Age, median (IQR) | 63 (54, 69) | 60 (51, 68) | 0.062 |
| AFP (ng/ml), median (IQR) | 9 (3, 54) | 35 (5.5, 1,795.5) | <0.001 |
| Albumin (g/dl), median (IQR) | 4 (3.5, 4.3) | 4 (3.5, 4.3) | 0.715 |

BAIAP2L2 in LIHC using HPA and discovered that BAIAP2L2 was overexpressed in liver cancer (Figure 5D). More importantly, qRT–PCR was conducted to evaluate gene expression, and we found that BAIAP2L2 mRNA expression was upregulated in a LIHC cell line (HEPG2) compared to a human normal liver cell line (LO2) ($p < 0.0001$) (Figure 5F).

## High BAIAP2L2 expression is correlated with clinical characteristics and pathological parameters in patients with LIHC

Clinical characteristics and gene expression data of 371 patients with LIHC were obtained from TCGA database. According to the mean value of BAIAP2L2, the patients with LIHC were divided into the high expression group and low expression group (Table 1), and then the Wilcoxon rank sum test and logistic regression were used to analyze the correlation between BAIAP2L2 expression and clinical features. High BAIAP2L2 expression was associated with histologic grade, pathologic stage, T stage, N stage, M stage, tumor status, Child-Pugh grade and residual tumor (Figure 5G). The results of univariate analysis using logistic regression demonstrated that BAIAP2L2 expression was connected with poor prognostic clinical characteristics in patients with LIHC (Table 2). High BAIAP2L2 expression was linked to sex [odds ratio (OR) = 2.149, 95% CI = 1.382–3.372, $p < 0.001$], age (OR = 0.662, 95% CI = 0.438–0.996, $p = 0.048$), histologic grade (G3&G4 vs. G1&G2: OR = 1.855, 95% CI = 1.208–2.867, $p = 0.005$) and AFP (OR = 2.544, 95% CI = 1.441–4.586, $p = 0.002$).

Additionally, a receiver operating characteristic (ROC) curve was carried out to fully evaluate the diagnostic value of BAIAP2L2 for LIHC. The area under the curve (AUC) of BAIAP2L2 was 0.891, which suggested high diagnostic value (Figure 6C). The time-dependent ROC curve demonstrated that BAIAP2L2 could accurately predict prognosis (Figure 6C). Moreover, univariate Cox analysis showed that

TABLE 2 BAIAP2L2 expression associated with clinicopathologic characteristics (logistic regression).

| Characteristics | Total (N) | Odds Ratio (OR) | *p*–value |
|---|---|---|---|
| Gender (Female vs. Male) | 371 | 2.149 (1.382–3.372) | <0.001 |
| Age (>60 vs. ≤60 years) | 370 | 0.662 (0.438–0.996) | 0.048 |
| T stage (T3 & T4 vs. T1 & T2) | 368 | 1.522 (0.949–2.459) | 0.083 |
| N stage (N1 vs. N0) | 256 | 3.048 (0.384–62.072) | 0.337 |
| M stage (M1 vs. M0) | 270 | 0.970 (0.115–8.184) | 0.976 |
| Pathologic stage (Stage III & Stage IV vs. Stage I & Stage II) | 347 | 1.514 (0.935–2.470) | 0.093 |
| Histologic grade (G3 & G4 vs. G1 & G2) | 366 | 1.855 (1.208–2.867) | 0.005 |
| Residual tumor (R2 & R1 vs. R0) | 342 | 1.611 (0.619–4.474) | 0.337 |
| Child–Pugh grade (B & C vs. A) | 239 | 0.766 (0.305–1.851) | 0.558 |
| Adjacent hepatic tissue inflammation (Mild & Severe vs. None) | 234 | 0.760 (0.454–1.270) | 0.296 |
| Vascular invasion (Yes vs. No) | 315 | 1.514 (0.951–2.423) | 0.082 |
| Fibrosis Ishak score (3/4 & 5/6 vs. 0 & 1/2) | 212 | 1.363 (0.793–2.353) | 0.264 |
| AFP (ng/ml) (>400 vs. ≤400) | 278 | 2.544 (1.441–4.586) | 0.002 |
| Albumin (g/dl) (≥3.5 vs. <3.5) | 297 | 0.965 (0.562–1.658) | 0.896 |

high BAIAP2L2 expression was dramatically correlated with poor OS [hazard ratio (HR) = 1.490, 95% CI =1.051–2.111, $p$ = 0.025] and DFS (HR = 1.603, 95% CI = 1.024–2.509, $p$ = 0.039) (**Figures 6A,B**). Eventually, a survival prediction nomogram using age, T stage, N stage, M stage, histologic grade and BAIAP2L2 was used to predict the 1-, 3-, and 5-year survival probability in LIHC (**Figure 6D**).

## BAIAP2L2 expression-correlated genes and proteins in LIHC

To further investigate the molecular mechanism of BAIAP2L2 in tumorigenesis, we attempted to screen out BAIAP2L2 expression-correlated genes and BAIAP2L2-binding proteins for a series of pathway enrichment analyses. The coexpression network of BAIAP2L2 was constructed by the LinkedOmics database. **Figure 7A** revealed the genes associated with BAIAP2L2 expression in the LIHC cohort. The 50 genes with the strongest positive and negative correlations are shown in **Figure 7B**. Then, GSEA was used to analyze the GO and KEGG enrichment analysis of genes coexpressed with BAIAP2L2. GO analysis revealed that genes coexpressed with BAIAP2L2 were mainly involved in chromosome localization and amine kinetochore organization (**Figure 7C**). KEGG pathway analysis showed that coexpressed genes were involved in fatty acid degradation and peroxisomes (**Figure 7D**). A PPI network of BAIAP2L2 was established (**Figure 7E**), showing that BAIAP2L2 interacts with MTSS1, AMPH, FCHO1, SYT9, PDK2, MTSS1L, PM20D1, CHST4 and PALM3. It has been reported that MTSS1 is a novel biomarker of tumor and elevated MTSS1 expression is associated with poor prognosis of liver cancer (40, 41).

## Immune infiltration analysis

Immune cells within the tumor microenvironment (TME) play important roles in tumorigenesis (42, 43). We used ssGSEA, TIMER, BioGPS and GEPIA to investigate the potential relationship between the infiltration level of different immune cells and BAIAP2L2 gene expression in LIHC. First, as shown in the BioGPS results in **Figure 8A**, higher expression of BAIAP2L2 was observed in B cells, dendritic cells (DCs), CD8+ T cells, CD4+ T cells, natural killer (NK) cells and monocytes. Meanwhile, we also observed that BAIAP2L2 was markedly overexpressed in liver tissue (**Figure 8A**). Then, we explored the association between BAIAP2L2 and the immune cell infiltration level quantified by ssGSEA in LIHC using Spearman correlation. The results showed that high BAIAP2L2 expression was positively correlated with the infiltration levels of T cells and NK cells (**Figure 8B**). The TIMER database further showed that the expression of BAIAP2L2 in LIHC was positively correlated with tumor infiltrating cells, including B cells, CD8+ T cells, CD4+ T cells, macrophages, and DCs (**Figure 8C**). Moreover, in addition to the correlation between BAIAP2L2 and the above immune infiltrating cells, we next sought to determine whether BAIAP2L2 was associated with the expression of more immune infiltrating cells by investigating related immune cell markers in LIHC in TIMER and GEPIA. The results showed that these immune cell markers were related to liver cancer, including B cells, CD8+ T cells, T follicular helper (Tfh) cells, T cells (general), Th1, Th2, Th9, Th17, Th22, Treg, exhausted T cells, M1 and M2 macrophages, tumor-associated macrophages (TAMs), monocytes, NK cells, neutrophils, and DCs (**Tables 3**, **4**).

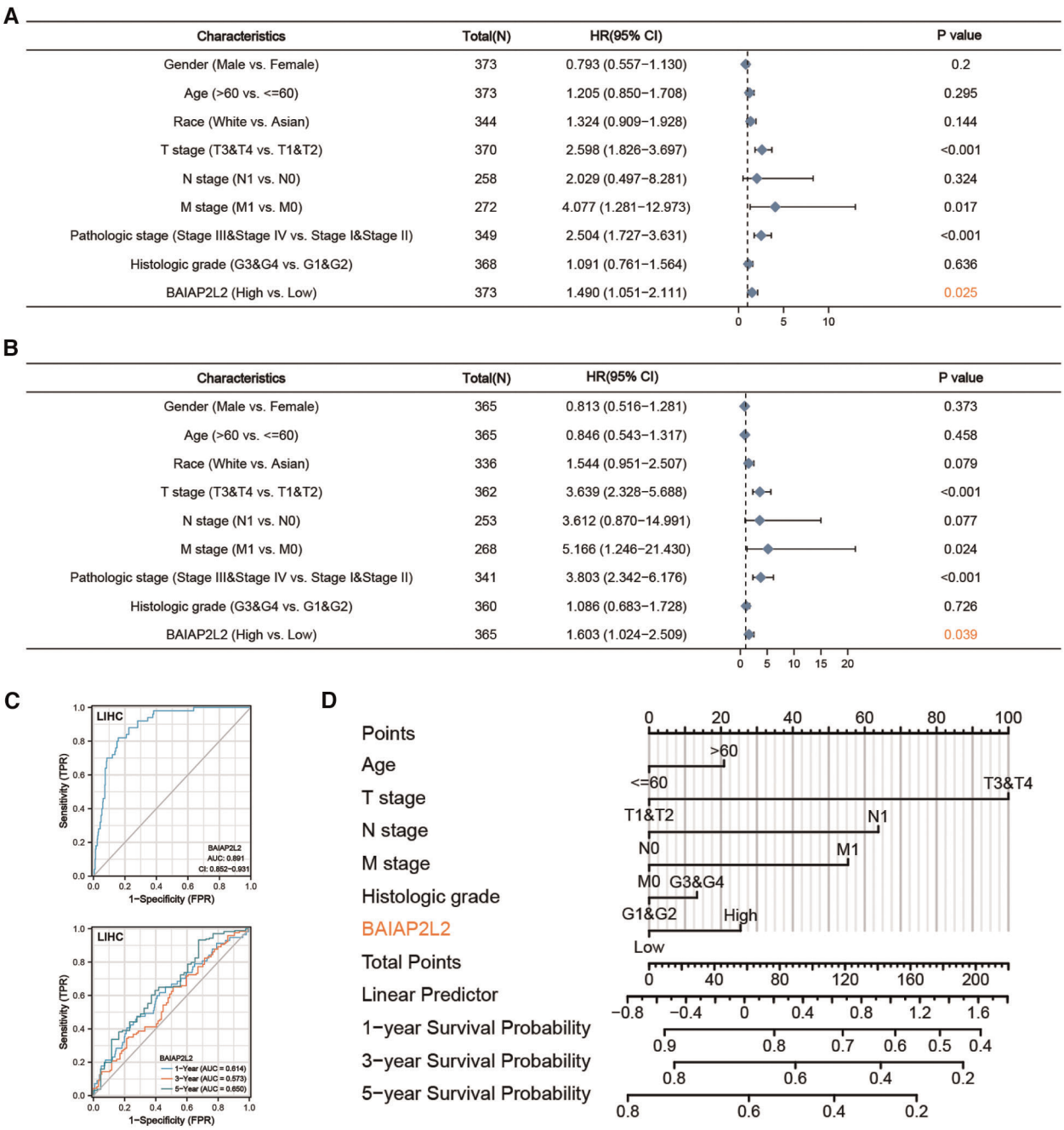


FIGURE 6
Forest plot, ROC curve and nomogram. (A) Forest plot of the Cox regression analysis in TCGA-LIHC (OS). (B) Forest plot of the Cox regression analysis in TCGA-LIHC (RFS). (C) ROC curve and time-dependent ROC curve for BAIAP2L2 in LIHC samples and adjacent Normal tissue samples from TCGA. (D) A nomogram for predicting the 1-, 3- and 5-year survival probability of patients.

## Discussion

In recent years, studies have suggested that BAIAP2L2 may be involved in the development of human cancer (36, 37, 44). However, the relationship between BAIAP2L2 and liver cancer has not been reported. Hence, we performed a comprehensive bioinformatics analysis of BAIAP2L2 expression and survival prognostic value in LIHC. Our results reveal for the first time that Overexpression of BAIAP2L2 is associated with poor prognosis of LIHC.

Our results showed that BAIAP2L2 was upregulated in BLCA, CHOL, ESCA, HNSC, KIRC, LIHC, LUAD, LUSC,

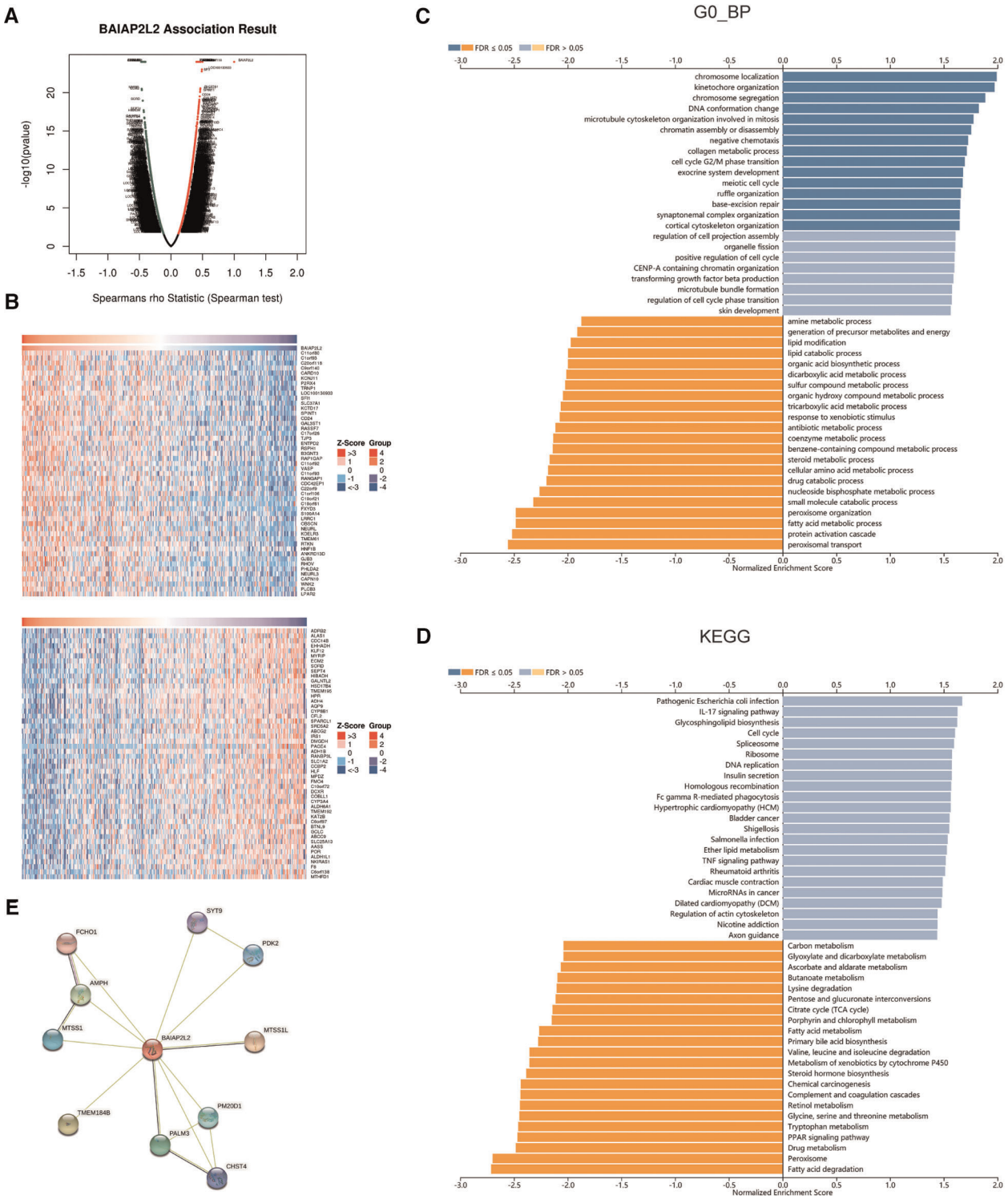


FIGURE 7
Genes and proteins coexpressed with BAIAP2L2 in LIHC from the LinkedOmics database. (**A**) Highly correlated genes identified by the Pearson test in the LIHC cohort. (**B**) Heat maps showing the top 50 genes positively and negatively correlated with BAIAP2L2 in LIHC (red: positively correlated genes; blue: negatively correlated genes). (**C**,**D**): Significantly enriched GO annotations and KEGG pathways of the genes coexpressed with BAIAP2L2 in LIHC. (**E**) A PPI network of BAIAP2L2 was generated using the STRING database.

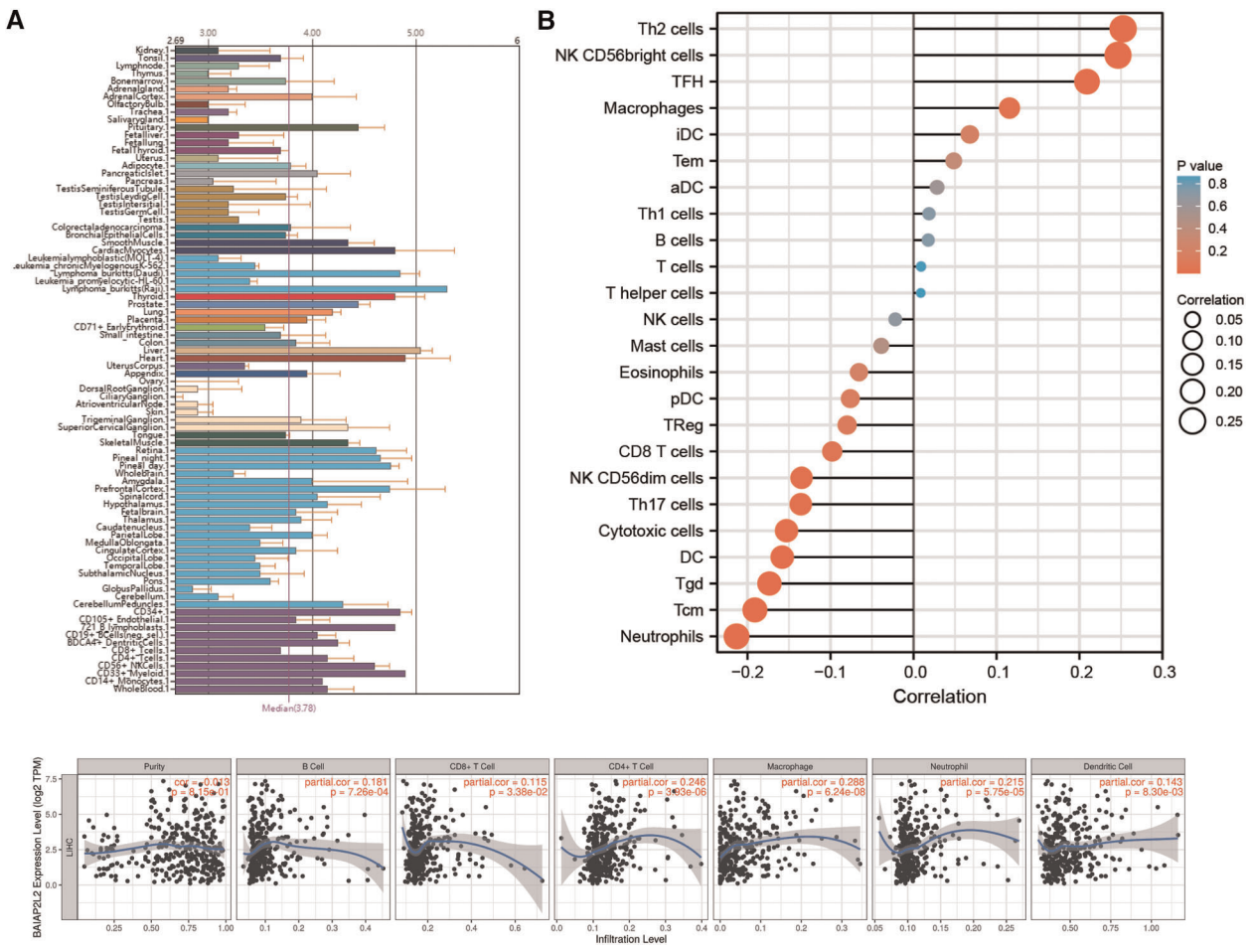


FIGURE 8
Correlation of BAIAP2L2 expression with the immune infiltration level. (A) Immune cell infiltration analysis of BAIAP2L2 in BioGPS. (B) The forest plot shows the correlation between BAIAP2L2 expression level and 24 immune cells. (C) BAIAP2L2 is significantly associated with tumor purity and is positively correlated with the infiltration of different immune cells using the TIMER database.

PAAD, PRAD and STAD based on TCGA. In 2020, Liu et al. found that BAIAP2L2 is highly expressed in STAD, and it can promote proliferation, migration and invasion and ultimately induce apoptosis of gastric cancer cells (20). BAIAP2L2 was also upregulated in PRAD, and it may promote tumorigenesis and malignant development (37). Upregulation of BAIAP2L2 was detected in various lung cancer cell lines and was deemed a novel biomarker and potential therapeutic target for LUAD (22). These reports support the results of this study. However, the relationship between BAIAP2L2 and LIHC has not been reported. Moreover, we found that BAIAP2L2 was significantly overexpressed in LIHC ($p < 0.001$). Interestingly, in combination with the survival analysis of the Kaplan–Meier Plotter and LinkedOmics databases, differences in both OS and DFS between the normal and tumor groups were observed only in LIHC. These results imply that BAIAP2L2 may play a unique and crucial role in LIHC.

Subsequently, we validated the expression of BAIAP2L2 mRNA and protein in LIHC using various online databases. All the results showed that BAIAP2L2 expression was upregulated in LIHC. To verify the above results, we performed qRT–PCR on HEPG2 cell and LO2 cell, and the results showed that the expression of BAIAP2L2 in HEPG2 cell was higher than that in LO2 cell.

LIHC is the fourth most common fatal malignancy and the sixth most common in terms of incidence cases in the world (3). The most common primary liver cancer, usually occurs in the context of chronic liver disease and is often diagnosed with liver cancer in advanced stages, resulting in its poor prognosis (45). It is of great significance to explore the pathogenesis of LIHC and identify potential molecular biomarkers. Therefore, we focused on the clinical significance and possible molecular mechanism of BAIAP2L2 in LIHC.

TABLE 3 Correlation analysis between BAIAP2L2 and gene markers of immune cells in TIMER (22).

| Cell type | Gene marker | LIHC ($n = 371$) | | | |
|---|---|---|---|---|---|
| | | None | | Purity | |
| | | cor | $p$ | cor | $p$ |
| B cell | CD19 | 0.115 | * | 0.117 | * |
| | CD21 (CR2) | 0.238 | **** | 0.257 | **** |
| | CD22 | 0.199 | *** | 0.18 | *** |
| T cell (general) | CD3D | 0.22 | **** | 0.24 | **** |
| | CD3E | 0.112 | * | 0.133 | * |
| | CD2 | 0.134 | ** | 0.153 | ** |
| Th1 | STAT4 | 0.181 | *** | 0.176 | ** |
| | STAT1 | 0.117 | * | 0.125 | * |
| | CD94 (KLRD1) | −0.115 | * | −0.117 | * |
| | IL12RB2 | −0.164 | ** | −0.164 | ** |
| | IL27RA | 0.324 | **** | 0.329 | **** |
| | TNF | 0.134 | ** | 0.154 | ** |
| Th2 | GATA3 | 0.135 | ** | 0.141 | ** |
| | CD184 (CXCR4) | 0.184 | *** | 0.182 | *** |
| Th9 | TGFBR2 | −0.139 | ** | −0.157 | ** |
| | IRF4 | 0.108 | * | 0.116 | * |
| | SPI1 | 0.196 | *** | 0.227 | **** |
| | TNF | 0.134 | ** | 0.154 | ** |
| Th17 | IL21R | 0.134 | ** | 0.144 | ** |
| Th22 | CCR10 | 0.237 | **** | 0.223 | **** |
| Treg | IL2RA | 0.138 | ** | 0.156 | ** |
| | CCR8 | 0.127 | * | 0.142 | ** |
| | TGFB1 | 0.332 | **** | 0.324 | **** |
| Exhausted T cell | PD-1 (PDCD1) | 0.173 | *** | 0.182 | *** |
| | TIM-3 (HAVCR2) | 0.192 | *** | 0.228 | **** |
| | CTLA4 | 0.172 | *** | 0.186 | *** |
| | LAG3 | 0.146 | ** | 0.136 | * |
| M1 Macrophage | IRF5 | 0.299 | **** | 0.308 | **** |
| | COX2 (PTGS2) | 0.141 | ** | 0.146 | ** |
| M2 Macrophage | ARG1 | −0.22 | **** | −0.205 | *** |
| | MRC1 | −0.157 | ** | −0.159 | ** |
| TAMs | CD80 | 0.125 | * | 0.151 | ** |
| | IL10 | 0.095 | 0.0665 | 0.106 | * |
| | CD68 | 0.138 | ** | 0.147 | ** |
| Monocyte | CD86 | 0.133 | * | 0.15 | ** |
| | CD14 | −0.193 | *** | −0.175 | ** |
| NK cell | NCAM1 | 0.177 | *** | 0.193 | *** |
| | CD94 (KLRD1) | −0.115 | * | −0.117 | * |
| | CD7 | 0.223 | **** | 0.235 | **** |
| Neutrophil | CD66b (CEACAM8) | 0.155 | ** | 0.176 | ** |
| | CD11b (ITGAM) | 0.159 | ** | 0.166 | ** |
| | CD15 (FUT4) | 0.389 | **** | 0.373 | **** |
| DCs | ITGAX | 0.198 | *** | 0.229 | **** |

*$p < 0.05$, **$p < 0.01$, ***$p < 0.001$, ****$p < 0.0001$.

Epigenetic changes have become an emerging applications for cancer biomarkers (46, 47). DNA methylation plays an important role in the development of cancer (48). Thus, we investigated whether the abnormal expression of BAIAP2L2 in cancer is related to epigenetics. According to **Figure 3**, we can see that high BAIAP2L2 expression was accompanied by gene alterations in LIHC. Furthermore, high BAIAP2L2 levels were

TABLE 4 Correlation analysis between BAIAP2L2 and gene markers of immune cells in GEPIA (31).

| Cell type | Gene marker | LIHC | | | |
|---|---|---|---|---|---|
| | | Tumor | | Normal | |
| | | R | $p$ | R | $p$ |
| B cell | CD19 | 0.15 | ** | 0.38 | ** |
| | CD20 (MS4A1) | 0.027 | 0.6 | 0.44 | ** |
| | CD21 (CR2) | 0.25 | **** | 0.31 | * |
| | CD22 | 0.22 | **** | 0.011 | 0.94 |
| | CD23 | −0.076 | 0.14 | 0.4 | ** |
| | CD24 | 0.5 | **** | 0.71 | **** |
| | CD40 | 0.024 | 0.64 | 0.32 | * |
| | CD72 | −0.004 | 0.94 | 0.39 | ** |
| | CD79a | 0.053 | 0.31 | 0.55 | **** |
| | CD138 | −0.0048 | 0.93 | 0.36 | * |
| CD8+ T cell | CD8A | 0.02 | 0.7 | 0.62 | **** |
| | CD8B | 0.02 | 0.7 | 0.61 | **** |
| Tfh | CXCR3 | 0.13 | ** | 0.55 | **** |
| | CXCR5 | 0.25 | **** | 0.2 | 0.16 |
| | ICOS | 0.11 | * | 0.54 | **** |
| T cell (general) | CD3D | 0.2 | *** | 0.56 | **** |
| | CD3E | 0.1 | * | 0.56 | **** |
| | CD2 | 0.12 | * | 0.52 | *** |
| Th1 | IFN-$\gamma$ (IFNG) | 0.02 | 0.7 | 0.38 | ** |
| | STAT4 | 0.18 | *** | 0.41 | ** |
| | STAT1 | 0.15 | ** | 0.53 | **** |
| | CD94 (KLRD1) | −0.077 | 0.14 | 0.33 | * |
| | IL12RB2 | −0.1 | * | 0.34 | * |
| | IL27RA | 0.33 | **** | 0.44 | ** |
| Th2 | GATA3 | 0.12 | * | 0.27 | 0.063 |
| | STAT6 | 0.14 | ** | 0.41 | ** |
| | CD184 (CXCR4) | 0.2 | *** | 0.57 | **** |
| | CD194 (CCR4) | 0.14 | ** | 0.41 | ** |
| Th9 | SPI1 | 0.21 | **** | 0.39 | ** |
| | TNF | 0.14 | ** | 0.35 | * |
| Th17 | IL21R | 0.13 | * | 0.47 | *** |
| | IL23R | 0.036 | 0.49 | 0.33 | * |
| | CD161 (KLRB1) | 0.042 | 0.42 | 0.29 | * |
| Th22 | CCR10 | 0.22 | **** | 0.28 | * |
| Treg | IL2RA | 0.15 | ** | 0.44 | ** |
| | FOXP3 | −0.0077 | 0.88 | 0.38 | ** |
| | CCR8 | 0.17 | ** | 0.23 | 0.12 |
| | CD127 (IL7R) | 0.1 | * | 0.55 | **** |
| | TGFB1 | 0.33 | **** | 0.5 | *** |
| Exhausted T cell | PD-1 (PDCD1) | 0.16 | ** | 0.67 | **** |
| | TIM-3 (HAVCR2) | 0.19 | *** | 0.31 | * |
| | CTLA4 | 0.14 | ** | 0.59 | **** |
| | LAG3 | 0.12 | * | 0.19 | 0.2 |
| M1 Macrophage | IRF5 | 0.33 | **** | 0.27 | 0.057 |
| | COX2 (PTGS2) | 0.15 | ** | 0.31 | * |
| | INOS (NOS2) | −0.019 | 0.71 | 0.46 | *** |
| M2 Macrophage | ARG1 | −0.18 | *** | 0.005 | 0.97 |
| | MRC1 | −0.13 | * | 0.086 | 0.55 |
| | VSIG4 | 0.064 | 0.22 | 0.29 | * |
| | MS4A4A | −0.011 | 0.84 | 0.32 | * |
| TAMs | CD80 | 0.13 | * | 0.34 | * |
| | IL10 | 0.074 | 0.16 | 0.35 | * |
| | CD68 | 0.14 | ** | 0.32 | * |
| Monocyte | CD86 | 0.14 | ** | 0.36 | * |

(*continued*)

TABLE 4 Continued

| Cell type | Gene marker | LIHC | | | |
|---|---|---|---|---|---|
| | | Tumor | | Normal | |
| | | R | *p* | R | *p* |
| | CD14 | −0.17 | ** | −0.16 | 0.28 |
| NK cell | NCAM1 | 0.16 | ** | 0.38 | ** |
| | CD94 (KLRD1) | −0.077 | 0.14 | 0.33 | 0.02 |
| | CD7 | 0.19 | *** | 0.37 | ** |
| Neutrophil | CD66b (CEACAM8) | 0.15 | ** | 0.47 | *** |
| | CD11b (ITGAM) | 0.16 | ** | 0.47 | *** |
| | CD15 (FUT4) | 0.41 | **** | 0.55 | **** |
| | CCR7 | 0.027 | 0.61 | 0.42 | ** |
| | MPO | −0.055 | 0.29 | 0.5 | *** |
| DCs | CD1C | 0.11 | * | 0.21 | 0.14 |
| | CD141 | 0.027 | 0.6 | 0.42 | ** |
| | HLA-DPB1 | 0.07 | 0.18 | 0.38 | ** |
| | HLA-DRA | 0.035 | 0.5 | 0.4 | ** |
| | THBD | 0.027 | 0.6 | 0.42 | ** |
| | NRP1 | 0.089 | 0.089 | 0.29 | * |
| | ITGAX | 0.21 | **** | 0.41 | ** |

*$p < 0.05$, **$p < 0.01$, ***$p < 0.001$, ****$p < 0.0001$.

associated with lower DNA methylation levels in LIHC. Results suggested that abnormally increased expression of BAIAP2L2 mRNA in LIHC may be the result of both genetic alterations and lower DNA methylation levels.

Additionally, we found that a high level of BAIAP2L2 indicated unfavorable survival probability in LIHC. Logistic regression analysis showed that high BAIAP2L2 expression was correlated with sex, age, histologic grade and alpha fetoprotein (AFP). It is well known that AFP is the most widely used serum biomarker for the diagnosis of primary liver cancer worldwide and is associated with poor prognosis (49). Simultaneously, Cox regression revealed that upregulation of BAIAP2L2 was an independent prognostic factor for poor prognosis of LIHC, along with pathological stage, T stage and distant metastasis (**Figure 6**). ROC analysis also indicated that BAIAP2L2 had a high diagnostic value in LIHC, and its AUC was 0.89. More importantly, a prognostic nomogram including age, T, M, N typing, histologic grade and BAIAP2L2 was constructed. The nomogram results suggested that BAIAP2L2 can reflect the prognosis of LIHC to some extent. These results demonstate that BAIAP2L2 plays an important role in the development of LIHC and may be an independent prognostic biomarker of LIHC.

To further investigate the molecular mechanism of the BAIAP2L2 gene in tumorigenesis, GSEA was used to analyze the GO and KEGG enrichment analysis of genes coexpressed with BAIAP2L2. The results suggest that MTSS1 has synergistic effect with BAIAP2L2. Huang et al. found that elevated MTSS1 expression is associated with poor prognosis of LIHC (41). In other words, MTSS1 and BAIAP2L2 may play a synergistic role in the carcinogenesis of LIHC.

In recent years, increasing evidence has shown that immune infiltration is closely related to malignant tumors (50–54). Therefore, we further analyzed the relationship between the carcinogenic effect of BAIAP2L2 and immune infiltration. Higher expression of BAIAP2L2 was observed in B cells, DCs, CD8+ T cells, CD4+ T cells, natural killer (NK) cells and monocytes in the BioGPS. Concurrently, the TIMER database showed that the expression of BAIAP2L2 in LIHC was positively correlated with tumor infiltrating cells, including B cells, DCs, CD8+ T cells, CD4+ T cells and macrophages. Moreover, LIHC was associated with immune cell markers, including B cells, CD8+ T cells, Tfh cells, T cells (general), Th1, Th2, Th9, Th17, Th22, Treg, exhausted T cells, M1 and M2 macrophages, TAMs, monocytes, NK cells, neutrophils, and DCs. Single-cell sequencing showed that CD8+ T cells were associated with liver cancer (55). Clinical samples also showed that immune cell markers were related to liver cancer, including B cells, Tfh cells, M1 macrophages, NK cells and neutrophils (56). Our findings are consistent with both studies. In summary, immune infiltration plays a crucial role in carcinogenesis.

Nevertheless, although we employed multiple bioinformatics databases to analyze the role of BAIAP2L2 in LIHC, this study still has some limitations. Firstly, although bioinformatics analysis is a powerful and efficient tool to help understand the molecular mechanisms and to identify potential biomarkers of LIHC, further experimental validations, such as evidence obtained from western blot and immunohistochemistry assays, are needed to confirm the prognosis value and immunological role of BAIAP2L2 in LIHC. Secondly, because most of the data come from public databases, there may be some biases caused by potential confounding factors. Finally, It appears that a single biomarker would lack enough prognosis power. Multiple biomarkers should be included to build a prognosis model to improve prognosis value. Unable to incorporate more hub genes is one of the limitations of our study. In future studies, we will try to combine hub gene to build a new prognosis model to improve specifificity and we will further validated in cell lines and animal models.

In conclusion, this is the first study to demonstrate the high expression of BAIAP2L2 and its prognostic value in LIHC. Our results also hinted at the potential role of BAIAP2L2 in modulating immune infiltration. These data provide a reference for future understanding of the role of BAIAP2L2 in LIHC.

## Conclusion

In summary, a pan-cancer analysis shows that BAIAP2L2 is highly expressed in LIHC and overexpression of BAIAP2L2 is associated with poor prognosis of LIHC. Furthermore,

BAIAP2L2 may be an independent prognostic biomarker of LIHC and be associated with immune infiltration. Nevertheless, the specific role and precise regulatory mechanism of BAIAP2L2 in LIHC need further far-ranging and thorough research.

## Data availability statement

The datasets presented in this study can be found in online repositories. The names of the repository/repositories and accession number(s) can be found below: The data can be accessed by following websites: https://cistrome.shinyapps.io/timer/, http://starbase.sysu.edu.cn/, http://ualcan.path.uab.edu/analysis.html, https://www.proteinatlas.org/, https://kmplot.com/analysis/, http://www.linkedomics.org/, https://string-db.org/, http://biogps.org/.

## Author contributions

XDH and WL performed the research and wrote the manuscript. YL took part in revising the article critically for important intellectual content. JXX conceived and designed the study. All authors contributed to the article and approved the submitted version.

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## Conflict of interest

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

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## References

1. Torre LA, Bray F, Siegel RL, Ferlay J, Lortet-Tieulent J, Jemal A. Global cancer statistics, 2012. *CA Cancer J Clin*. (2015) 65(2):87–108. doi: 10.3322/caac.21262

2. Bray F, Ferlay J, Soerjomataram I, Siegel RL, Torre LA, Jemal A. Global cancer statistics 2018: GLOBOCAN estimates of incidence and mortality worldwide for 36 cancers in 185 countries. *CA Cancer J Clin*. (2018) 68(6):394–424. doi: 10.3322/caac.21492

3. Villanueva A. Hepatocellular carcinoma. *N Engl J Med*. (2019) 380 (15):1450–62. doi: 10.1056/NEJMra1713263

4. Melaram R. Environmental risk factors implicated in liver disease: a mini-review. *Front Public Health*. (2021):9–683719.

5. Kudo M, Kawamura Y, Hasegawa K, Tateishi R, Kariyama K, Shiina S, et al. Management of hepatocellular carcinoma in Japan: JSH consensus statements and recommendations 2021 update. *Liver Cancer*. (2021) 10(3):181–223. doi: 10.1159/000514174

6. Chen S, Cao Q, Wen W, Wang H. Targeted therapy for hepatocellular carcinoma: challenges and opportunities. *Cancer Lett*. (2019) 460:1–9.

7. Kubli SP, Berger T, Araujo DV, Siu LL, Mak TW. Beyond immune checkpoint blockade: emerging immunological strategies. *Nat Rev Drug Discov*. (2021 Dec) 20 (12):899–919. doi: 10.1038/s41573-021-00155-y

8. Pykäläinen A, Boczkowska M, Zhao H, Saarikangas J, Rebowski G, Jansen M, et al. Pinkbar is an epithelial-specific BAR domain protein that generates planar membrane structures. *Nat Struct Mol Biol*. (2011) 18(8):902–7. doi: 10.1038/nsmb.2079

9. Ahmed S, Goh WI, Bu W. I-BAR domains, IRSp53 and filopodium formation. *Semin Cell Dev Biol*. (2010) 21(4):350–6. doi: 10.1016/j.semcdb.2009.11.008

10. Zhao H, Pykäläinen A, Lappalainen P. I-BAR domain proteins: linking actin and plasma membrane dynamics. *Curr Opin Cell Biol*. (2011) 23(1):14–21. doi: 10.1016/j.ceb.2010.10.005

11. Chen Y, Aardema J, Misra A, Corey SJ. BAR Proteins in cancer and blood disorders. *Int J Biochem Mol Biol*. (2012) 3(2):198–208.

12. Sudhaharan T, Hariharan S, Lim JSY, Liu JZ, Koon YL, Wright GD, et al. Superresolution microscopy reveals distinct localisation of full length IRSp53 and its I-BAR domain protein within filopodia. *Sci Rep*. (2019) 9(1):2524. doi: 10.1038/s41598-019-38851-w

13. Huang LY, Wang X, Cui XF, Li H, Zhao J, Wu CC, et al. IRTKS Is correlated with progression and survival time of patients with gastric cancer. *Gut*. (2018) 67 (8):1400–9. doi: 10.1136/gutjnl-2016-313478

14. Chao A, Tsai CL, Jung SM, Chuang WC, Kao C, Hsu A, et al. BAI1-associated protein 2-like 1 (BAIAP2L1) is a potential biomarker in ovarian cancer. *PLoS One*. (2015) 10(7):e0133081–00. doi: 10.1371/journal.pone.0133081

15. Funato Y, Terabayashi T, Suenaga N, Seiki M, Takenawa T, Miki H. IRSp53/Eps8 complex is important for positive regulation of Rac and cancer cell motility/invasiveness. *Cancer Res*. (2004) 64(15):5237–44. doi: 10.1158/0008-5472.CAN-04-0327

16. Antoine M, Vandenbroere I, Ghosh S, Erneux C, Pirson I. IRSp53 is a novel interactor of SHIP2: a role of the actin binding protein Mena in their cellular localization in breast cancer cells. *Cell Signal*. (2020) 73:109692. doi: 10.1016/j.cellsig.2020.109692

17. Liu K, Jiao XD, Hao JL, Qin BD, Wu Y, Chen W, et al. MTSS1 Inhibits metastatic potential and induces G2/M phase cell cycle arrest in gastric cancer. *Onco Targets Ther*. (2019) 12:5143–52. doi: 10.2147/OTT.S203165

18. Chen J, Huang L, Zhu Q, Wang Z, Tang Z. MTSS1 Hypermethylation is associated with prostate cancer progression. *J Cell Physiol*. (2020) 235 (3):2687–97. doi: 10.1002/jcp.29172

19. Guo H, Peng J, Hu J, Chang S, Liu H, Luo H, et al. BAIAP2L2 Promotes the proliferation, migration and invasion of osteosarcoma associated with the Wnt/β-catenin pathway. *J Bone Oncol*. (2021) 31:100393. doi: 10.1016/j.jbo.2021.100393

20. Liu J, Shangguan Y, Sun J, Cong W, Xie Y. BAIAP2L2 Promotes the progression of gastric cancer via AKT/mTOR and Wnt3a/β-catenin signaling pathways. *Biomed Pharmacother*. (2020) 129:110414. doi: 10.1016/j.biopha.2020.110414

21. Liu S, Wang W, Zhao Y, Liang K, Huang Y. Identification of potential key genes for pathogenesis and prognosis in prostate cancer by integrated analysis of gene expression profiles and the cancer genome atlas. *Front Oncol*. (2020) 10:809. doi: 10.3389/fonc.2020.00809

22. Xu L, Du H, Zhang Q, Wang C, Yan L, Tian G, et al. BAI1-associated Protein 2-like 2 is a potential biomarker in lung cancer. *Oncol Rep*. (2019) 41 (2):1304–12.

23. Li T, Fan J, Wang B, Traugh N, Chen Q, Liu JS, et al. TIMER: a web server for comprehensive analysis of tumor-infiltrating immune cells. *Cancer Res*. (2017) 77(21):e108–10. doi: 10.1158/0008-5472.CAN-17-0307

24. Li T, Fu J, Zeng Z, Cohen D, Li J, Chen Q, et al. TIMER2.0 For analysis of tumor-infiltrating immune cells. *Nucleic Acids Res*. (2020) 48(W1):W509–14. doi: 10.1093/nar/gkaa407

25. Li JH, Liu S, Zhou H, Qu LH, Yang JH. Starbase v2.0: decoding miRNA-ceRNA, miRNA-ncRNA and protein-RNA interaction networks from large-scale CLIP-Seq data. *Nucleic Acids Res*. (2014) 42(Database issue):D92–7.

26. Chandrashekar DS, Bashel B, Balasubramanya SAH, Creighton CJ, Ponce-Rodriguez I, Chakravarthi BVSK, et al. UALCAN: a portal for facilitating tumor subgroup gene expression and survival analyses. *Neoplasia (New York, N.Y.)*. (2017) 19(8):649–58. doi: 10.1016/j.neo.2017.05.002

27. Uhlen M, Zhang C, Lee S, Sjöstedt E, Fagerberg L, Bidkhori G, et al. A pathology atlas of the human cancer transcriptome. *Science (New York, N.Y.)*. (2017) 357:6352. doi: 10.1126/science.aan2507

28. Lanczky A, Gyorffy B. Web-based survival analysis tool tailored for medical research (KMplot): development and implementation. *J Med Internet Res*. (2021) 23(7):e27633. doi: 10.2196/27633

29. Vasaikar SV, Straub P, Wang J, Zhang B. Linkedomics: analyzing multi-omics data within and across 32 cancer types. *Nucleic Acids Res*. (2018) 46(D1): D956–63. doi: 10.1093/nar/gkx1090

30. Cerami E, Gao J, Dogrusoz U, Gross BE, Sumer SO, Aksoy BA, et al. The cBio cancer genomics portal: an open platform for exploring multidimensional cancer genomics data. *Cancer Discov*. (2012) 2(5):401–4. doi: 10.1158/2159-8290.CD-12-0095

31. Gao J, Aksoy BA, Dogrusoz U, Dresdner G, Gross B, Sumer SO, et al. Integrative analysis of complex cancer genomics and clinical profiles using the cBioPortal. *Sci Signal*. (2013) 6(269):l1.

32. Chandrashekar DS, Karthikeyan SK, Korla PK, Patel H, Shovon AR, Athar M, et al. UALCAN: an update to the integrated cancer data analysis platform. *Neoplasia*. (2022 Mar) 25:18–27. doi: 10.1016/j.neo.2022.01.001

33. Szklarczyk D, Gable AL, Lyon D, Junge A, Wyder S, Huerta-Cepas J, et al. STRING V11: protein-protein association networks with increased coverage, supporting functional discovery in genome-wide experimental datasets. *Nucleic Acids Res*. (2019) 47(D1):D607–13. doi: 10.1093/nar/gky1131

34. Tang Z, Li C, Kang B, Gao G, Li C, Zhang Z. GEPIA: a web server for cancer and Normal gene expression profiling and interactive analyses. *Nucleic Acids Res*. (2017) 45(W1):W98–W102. doi: 10.1093/nar/gkx247

35. Wu C, Jin X, Tsueng G, Afrasiabi C, Su AI. BioGPS: building your own mash-up of gene annotations and expression profiles. *Nucleic Acids Res*. (2016) 44(D1):D313–6. doi: 10.1093/nar/gkv1104

36. Hu W, Wang G, Yarmus LB, Wan Y. Combined methylome and transcriptome analyses reveals potential therapeutic targets for EGFR wild type lung cancers with low PD-L1 expression. *Cancers (Basel)*. (2020) 12(9):2496. doi: 10.3390/cancers12092496

37. Song Y, Zhuang G, Li J, Zhang M. BAIAP2L2 Facilitates the malignancy of prostate cancer (PCa) via VEGF and apoptosis signaling pathways. *Genes Genomics*. (2021) 43(4):421–32. doi: 10.1007/s13258-021-01061-8

38. Villanueva L, Álvarez-Errico D, Esteller M. The contribution of epigenetics to cancer immunotherapy. *Trends Immunol*. (2020) 41(8):676–91. doi: 10.1016/j.it.2020.06.002

39. Skvortsova K, Stirzaker C, Taberlay P. The DNA methylation landscape in cancer. *Essays Biochem*. (2019) 63(6):797–811. doi: 10.1042/EBC20190037

40. Fan H, Chen L, Zhang F, Quan Y, Su X, Qiu X, et al. MTSS1, A novel target of DNA methyltransferase 3B, functions as a tumor suppressor in hepatocellular carcinoma. *Oncogene*. (2012) 31(18):2298–308. doi: 10.1038/onc.2011.411

41. Huang XY, Huang ZL, Xu B, Chen Z, Re TJ, Zheng Q, et al. Elevated MTSS1 expression associated with metastasis and poor prognosis of residual hepatitis B-related hepatocellular carcinoma. *J Exp Clin Cancer Res CR*. (2016) 35(1):85. doi: 10.1186/s13046-016-0361-8

42. Lei X, Lei Y, Li JK, Du WX, Li RG, Yang J, et al. Immune cells within the tumor microenvironment: biological functions and roles in cancer immunotherapy. *Cancer Lett*. (2020) 470:126–33. doi: 10.1016/j.canlet.2019.11.009

43. Wu SY, Liao P, Yan LY, Zhao QY, Xie ZY, Dong J, et al. Correlation of MKI67 with prognosis, immune infiltration, and T cell exhaustion in hepatocellular carcinoma. *BMC Gastroenterol*. (2021) 21(1):416. doi: 10.1186/s12876-021-01984-2

44. Carlton AJ, Halford J, Underhill A, Jeng JY, Avenarius MR, Gilbert ML, et al. Loss of Baiap2l2 destabilizes the transducing stereocilia of cochlear hair cells and leads to deafness. *J Physiol (Lond)*. (2021) 599(4):1173–98. doi: 10.1113/JP280670

45. Craig AJ, von Felden J, Garcia-Lezana T, Sarcognato S, Villanueva A. Tumour evolution in hepatocellular carcinoma. *Nat Rev Gastroenterol Hepatol*. (2020) 17(3):139–52. doi: 10.1038/s41575-019-0229-4

46. Grady WM, Yu M, Markowitz SD. Epigenetic alterations in the gastrointestinal tract: current and emerging use for biomarkers of cancer. *Gastroenterology*. (2021) 160(3):690–709. doi: 10.1053/j.gastro.2020.09.058

47. Jung G, Hernández-Illán E, Moreira L, Balaguer F, Goel A. Epigenetics of colorectal cancer: biomarker and therapeutic potential. *Nat Rev Gastroenterol Hepatol*. (2020) 17(2):111–30. doi: 10.1038/s41575-019-0230-y

48. Dawson MA, Kouzarides T. Cancer epigenetics: from mechanism to therapy. *Cell*. (2012) 150(1):12–27. doi: 10.1016/j.cell.2012.06.013

49. Chen T, Dai X, Dai J, Ding C, Zhang Z, Lin Z, et al. AFP Promotes HCC progression by suppressing the HuR-mediated Fas/FADD apoptotic pathway. *Cell Death Dis*. (2020) 11(10):822. doi: 10.1038/s41419-020-03030-7

50. Picard E, Verschoor CP, Ma GW, Pawelec G. Relationships between immune landscapes, genetic subtypes and responses to immunotherapy in colorectal cancer. *Front Immunol*. (2020) 11:369. doi: 10.3389/fimmu.2020.00369

51. Rosenthal R, Cadieux EL, Salgado R, Bakir MA, Moore DA, Hiley CT, et al. Neoantigen-directed immune escape in lung cancer evolution. *Nature*. (2019) 567 (7749):479–85. doi: 10.1038/s41586-019-1032-7

52. Singh AK, McGuirk JP. CAR T cells: continuation in a revolution of immunotherapy. *Lancet Oncol*. (2020) 21(3):e168–78. doi: 10.1016/S1470-2045(19)30823-X

53. Moral JA, Leung J, Rojas LA, Ruan J, Zhao J, Sethna Z, et al. ILC2s Amplify PD-1 blockade by activating tissue-specific cancer immunity. *Nature*. (2020) 579 (7797):130–5. doi: 10.1038/s41586-020-2015-4

54. Yan Y, Nie K, Zheng J, Jiang X, Huang Y, Zheng Z, et al. High endothelin receptor type A expression as an independent prognostic biomarker and correlated with immune infiltrates in stomach adenocarcinoma. *Cancer Manag Res*. (2021) 13:5013–26. doi: 10.2147/CMAR.S313078

55. Zheng C, Zheng L, Yoo JK, Guo H, Zhang Y, Guo X, et al. Landscape of infiltrating T cells in liver cancer revealed by single-cell sequencing. *Cell*. (2017) 169(7):1342–1356.e16. doi: 10.1016/j.cell.2017.05.035

56. Rohr-Udilova N, Klinglmüller F, Schulte-Hermann R, Stift J, Herac M, Salzmann M, et al. Deviations of the immune cell landscape between healthy liver and hepatocellular carcinoma. *Sci Rep*. (2018) 8(1):6220. doi: 10.1038/s41598-018-24437-5

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# The cause analysis of benign uretero-ileal anastomotic stricture after radical cystectomy and urinary diversion

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**Background:** Benign uretero-ileal anastomotic stricture (UIAS) is a major complication following radical cystectomy (RC) and ileal orthotopic bladder substitution, and it can occur in combination with other complications. But risk factors for patients with UIAS have not been well described.

**Material and methods:** We retrospectively reviewed 198 patients treated with RC for bladder cancer from 2014 to 2019 at the Zhejiang Provincial People's Hospital. Patient demographic and clinical variables were examined to determine the risk factors associated with UIAS by univariate and multivariate logistic regression analysis.

**Results:** A total of 180 patients into the group standards and in all 360 uretero-ileal anastomoses. Among the above cases, 22 patients developed UIAS, including 10 cases of left UIAS, nine cases of right UIAS, and three cases of bilateral UIAS. There was no difference in demographic, operative, or perioperative variables between patients with and without UIAS. In a multivariate analysis, after adjusting for gender, age, surgical methods, and underlying diseases, intraoperative or postoperative blood transfusion (HR = 0.144, P <0.01), postoperative urinary tract infection (HR = 3.624, P <0.01), and extracorporeal bladder anastomosis (HR = 3.395, P = 0.02) significantly increased the risk of UIAS.

**Conclusions:** In our experience, intraoperative or postoperative blood transfusion, postoperative urinary tract infection, and extracorporeal neobladder anastomoses increased the risk of UIAS after radical cystectomy and ileal orthotopic bladder substitution surgery. Further studies with larger samples are necessary to validate this result.

KEYWORDS

**bladder cancer, urinary diversion, anastomosis, radical cystectomy, uretero-ileal anastomotic stricture**

## Introduction

Bladder cancer was the ninth most common cancer worldwide and the 13th most common cause of death, according to the latest research data. In recent years, the incidence of bladder cancer has been increasing year by year (1). The main performance is male incidence rate is higher than female, rural incidence rate is higher than city (1). Diagnosis, treatment, and 5-year survival rates for bladder cancer have remained largely unchanged since the 1990s. Radical cystectomy (RC) with urinary diversion (UD) is standard therapy for muscle-invasive bladder cancer and high-risk non-muscle-invasive disease (2). Most strictures occur between 6 months and 18 months after surgery. The overall complication rate is reported to be as high as 25%–35% (3). The published rate of UIAS after urinary diversion has a varying incidence in the literature (4). The incidence of postoperative benign UIAS has been reported to be 1% to 30% and varies considerably. Besides, complications after ileal bladder surgery are mainly urinary tract infection, incision infection, uretero-ileum stricture, intestinal fistula, intestinal obstruction, etc. (5, 6). Among them, urinary tract infections are the most common. If the ureteral outlet stricture is not treated in time, it is prone to complicated upper urinary tract infections, stones, renal insufficiency, and other serious complications. It has been proposed that the cause of UIAS is likely multifactorial. The causes of UIAS include anastomotic fibrosis, inflammation, and tumor recurrence, among which fibrosis is the most common factor (7). Excessive dissection and freeing of the ureter may lead to ureteral damage and ischemia, followed by inflammation, fibrosis, and scar formation (7). We retrospectively analyzed our surgical experience and demographic data in order to determine the risk factors for the formation of UIAS.

## Materials and methods

We obtained approval from our institutional review board before initiating this analysis. A retrospective database of all patients who underwent RC with an ileal conduit or an ileal orthotopic neobladder for bladder cancer at Zhejiang Provincial People's Hospital from 2014 to 2019 was analyzed. Patients' clinical characteristics were retrieved from hospital archives, including gender, age, body mass index, the American Society of Anesthesiologists (ASA), comorbidities, chemotherapy history, drinking history, and smoking history. Preoperative data: hemoglobin, WBC level, creatinine, albumin, uric acid level, alanine transferase, aspartate transferase, etc. Intraoperative data included intraoperative blood transfusion, lymph node dissection, the duration of the operation, and the preparation of a new bladder. Postoperative: hemoglobin, creatinine, postoperative urinary tract infection, average length of hospital stay. The diagnosis of UIAS is mainly made through radiography, enhanced CT, and three-dimensional reconstruction of the urinary system. If there is clear radiologic evidence, UIAS has occurred. Malignant strictures were excluded from the study. An enhanced CT scan of the urinary system would be performed every 3 months within 1 year after surgery, every 6 months within 1–5 years, and annually after 5 years. The shortest observation period is 1 year.

All the risk variables with UIAS were analyzed as a binary variable and assessed by univariable logistic regression analysis to detect outcome variables with significant odds ratios.

The variables that attained significance in univariate analysis included intraoperative or postoperative blood transfusion, extracorporeal neobladder anastomoses, and postoperative urinary tract infection. They were entered into a multivariable logistic model with an interaction term to ascertain whether they retained significance. All statistical tests were implemented at $p \leq 0.05$ significance level.

## Results

A total of 198 patients with radical bladder cancer were included in the study. Cases, which occur Malignant UIAS, with other Malignant tumor, loss to follow-up nine patients, finally only 180 patients into the group standards. The average postoperative hospital stay was 14.4 days. Every UIAS is a unit at risk of developing stricture and in all 360 uretero-ileal anastomosis, stricture patients a total of 22 cases, 10 cases were on the left side of the narrow, the right side of the narrow nine cases, bilateral stricture (three cases), stricture rate was 6.9%. Demographic information and clinical data collected are shown in Tables 1, 2.

Of 180 patients, the median age was 67 years, with 159 (88.3%) males. A total of 60 (33.3%) cases were occurred extracorporeal neobladder anastomoses, 22 (12.2%) patients were undergone intraoperative or postoperative blood transfusion and 40 (22.2%) patients were developed urinary tract infection. The incidence of bilateral strictures was 13.6%. Left side stricture was 45.5%, while the right side was 40.9%. The median interval from postoperative to diagnosis of stricture was 11 months. Univariate logistic regression analysis with narrow correlations between each variable is summarized in Figure 1.

Multivariate Logistic regression analysis was performed on UIAS risk factors with statistical differences ($P < 0.05$) in

TABLE 1 Summary of continuous variables collected.

| variables | Minimum | Median | Maximum |
|---|---|---|---|
| **Age** | 43 | 67 | 92 |
| **BMI** | 16 | 22 | 28 |
| **ASA score** | 1 | 2 | 3 |

TABLE 2 Summary of categorical variables recorded.

| Variables | N |
|---|---|
| Age | |
| 43–67 | 93 |
| 67–92 | 87 |
| BMI | |
| Normal | 110 |
| Abnormal | 70 |
| ASA score | |
| **1** | 116 |
| **2** | 41 |
| **3** | 23 |
| Sex | |
| Male | 159 |
| Female | 21 |
| Diabetic | |
| Yes | 154 |
| No | 26 |
| Smoking | |
| Yes | 102 |
| No | 78 |
| Drinking | |
| Yes | 50 |
| No | 130 |
| Preoperative HB | |
| ≥120 g/L | 155 |
| <120 g/L | 25 |
| Preoperative WBC | |
| >10 × 109/L | 13 |
| ≤10 × 109/L | 167 |
| Preoperative albumin | |
| ≥40 g/L | 106 |
| <40 g/L | 74 |
| Preoperative Uric acid level | |
| >357 umol/L | 56 |
| ≤357 umol/L | 124 |
| Preoperative Urea level | |
| >8.8 mmol/L | 39 |
| ≤8.8 mmol/L | 141 |
| Preoperative Creatinine | |
| >123 umol/L | 20 |
| ≤123 umol/L | 160 |
| Preoperative ALT | |
| >40 U/L | 18 |
| ≤40 U/L | 162 |
| Preoperative AST | |
| >35 U/L | 15 |
| ≤35 U/L | 165 |
| Preoperative cruenturesis | |
| Yes | 173 |
| No | 7 |
| Urine microscopy for white blood cells | |
| Yes | 119 |
| No | 61 |
| Blood transfusion | |
| Yes | 22 |
| No | 158 |
| Bricker anastomotic technique | |
| Yes | 101 |
| No | 79 |
| Intraoperative bleeding>400 ml | |
| Yes | 79 |
| No | 101 |
| Running Anastomosis | |
| Yes | 145 |
| No | 35 |
| Operation time>4 h | |
| Yes | 141 |
| No | 39 |
| Robot assisted surgery | |
| Yes | 105 |
| No | 75 |
| Intracorporeal neobladder anastomoses | |
| Yes | 120 |
| No | 60 |
| Postoperative HB (g/L) | |
| ≥120 g/L | 109 |
| <120 g/L | 71 |
| Postoperative WBC | |
| >10 × 109/L | 108 |
| ≤10 × 109/L | 72 |
| Postoperative Creatinine | |
| >123 umol/L | 20 |
| ≤123 umol/L | 160 |
| Postoperative urinary tract infection | |
| Yes | 40 |
| No | 140 |

*(Continued)*

univariate analysis. Intraoperative or postoperative blood transfusion (HR = 0.144, P <0.01), postoperative urinary tract infection (HR = 3.624, P <0.01), and extracorporeal bladder anastomosis (HR = 3.395, P = 0.02) were the variables most strongly associated with UIAS. Besides, the results of three variables that were taken into a multivariable logistic regression analysis are shown in Table 3.

Nomogram based on a logistic regression model: Based on the R software, assign values to the inclusion indicators of patients with UIAS and use the regression coefficient values

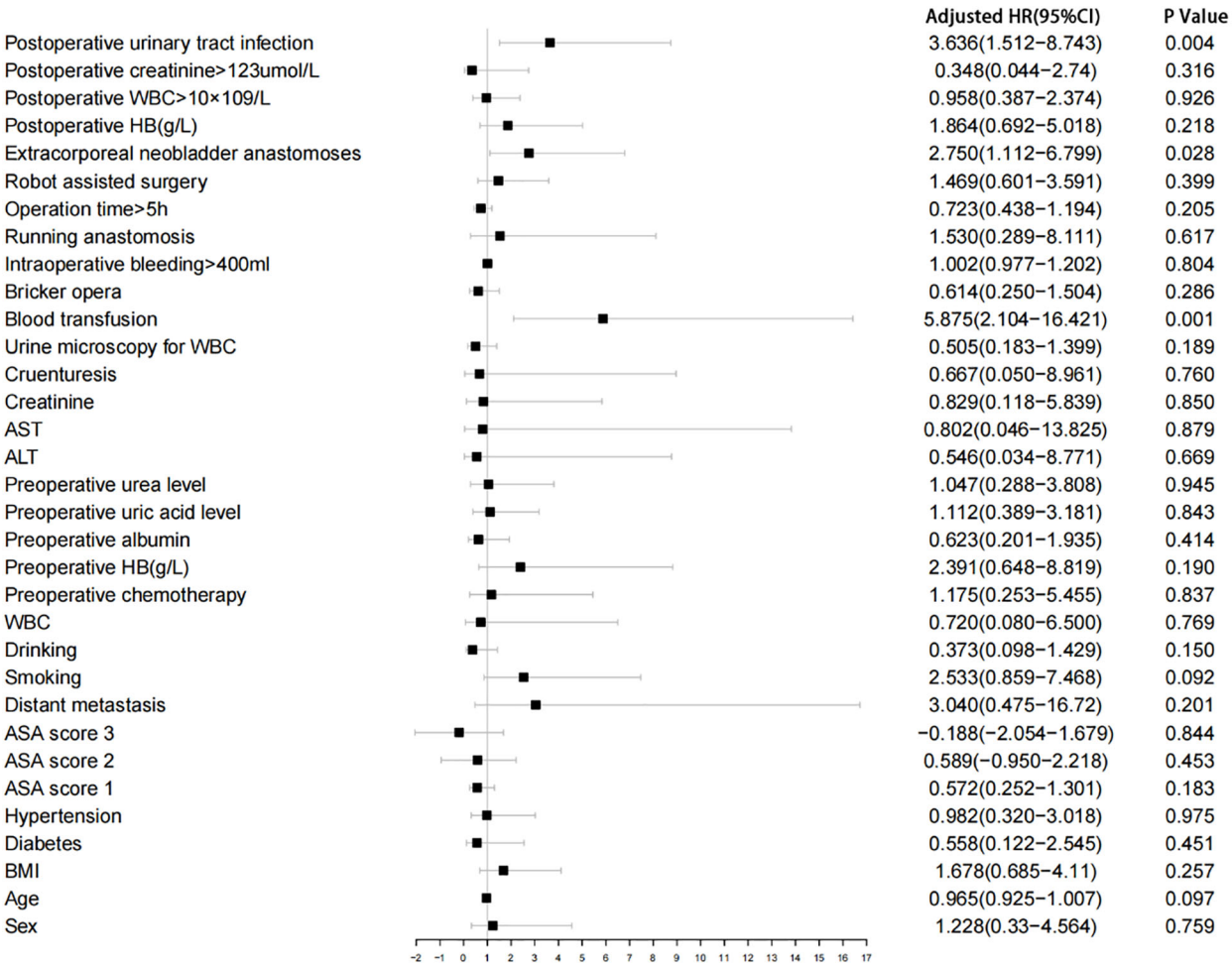


FIGURE 1
Univariate logistic regression analysis. WBC, white blood cell; ALT, Alanine transferase; AST, Aspartate transferase; HB, Hemoglobin.

corresponding to the three statistically significant indicators in the logistic regression analysis and the R language software program to obtain the score corresponding to each indicator, and then draw a list of items predicting the occurrence of bacterial infections. In the nomogram, sum the scores corresponding to each indicator and find the corresponding UISA probability through the total score. The results are shown in Figure 2.

TABLE 3 Multivariable Logistic regression analysis.

| Multivariate analysis | HR | P |
|---|---|---|
| Blood transfusion | | |
| No | 1 | |
| Yes | 0.144 (0.046–0.451) | 0.001 |
| Bladder anastomosis | | |
| Intra | 1 | |
| extra | 3.395 (1.241–9.283) | 0.017 |
| Urinary tract infection | | |
| No | 1 | |
| Yes | 3.624 (1.41–9.31) | 0.007 |

In the model fitted this time, the likelihood ratio test result of whether all parameters are 0 is P <0.05, that is, the OR value of at least one variable among the included variables is statistically significant, and the model is generally meaningful. The P-value of the goodness of fit is not less than the test level (p >0.05), the information in the current data has been fully extracted, and the goodness of fit of the model is high. This study found that anastomosing a new bladder in the extracorporeal setting compared to anastomosing a new bladder in the intracorporeal setting has an increased risk of UIAS. Blood transfusions during or after surgery also increased the risk of UIAS. At the same time, consistent with other related research, postoperative urinary tract infection was also associated with an increased risk of UIAS. As the risk factors of patients increase,

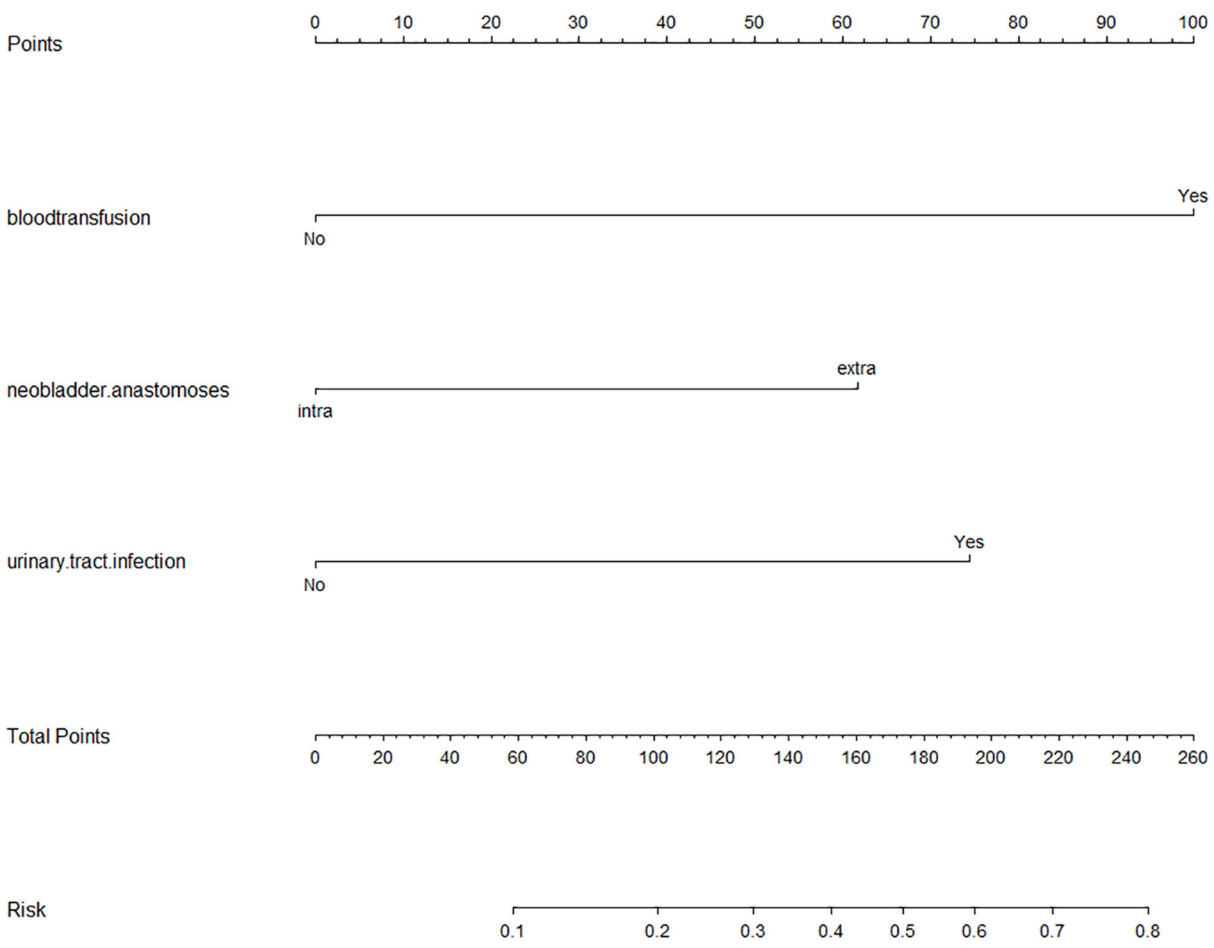


FIGURE 2
Nomogram based on Logistic regression model. Abbreviations: HR, hazard ratio. The scores of the three risk factors are added together to form the total score, ranging from 0 to 260 points. The percentage corresponding to the total score downward is the probability of UIAS in patients with Radical cystectomy with urinary diversion.

the proportion of UISA has a strong tendency to increase. At least one risk factor has a strong predictive effect on the development of UIAS.

## Discussion

RC and standard pelvic lymph node dissection: Bilateral pelvic lymph nodes were exposed and dissected. Free ureter from the ureteral wall segment. The peritoneum and vascular sheath were opened, and the internal and external iliac and obturator lymph nodes were dissected. Free cystorectal space and anterior bladder space. The bladder and prostatic ligaments were dealt with. Treatment of the prostate and urethra: the prostate and bladder are completely removed. Preparation of a new bladder: A mesangial enteral loop was taken about 20–30 cm from 10 to 20 cm away from the ileocecal part, and the broken ends of the proximal and distal ileocecal parts were anastomosed end to end or side to side, and the mesangial was sutured.

Furthermore, the following points should receive special attention in the operation: 1. minimize free tissue around the ureter to reduce blood supply; 2. avoid clamping the broken end of the ureter; 3. Ureteral length is retained to avoid excessive ureteral tension; and 4. ureteral stents are implanted to prevent stenosis.

Several retrospective studies have been conducted to identify risk factors associated with UIAS after radical cystectomy and ileal conduit formation. Current studies suggest that postoperative anastomotic leakage, postoperative urinary tract infection, and anastomotic ischemia would increase UIAS (8). In our study, intraoperative blood loss was not well reflected due to the subjective evaluation of intraoperative bleeding, intraoperative

and postoperative blood transfusion patients had greater intraoperative bleeding, which may lead to anastomotic ischemia and cause anastomotic stricture. Postoperative urinary tract infection also increased the risk of anastomotic stricture, which may be related to delayed wound healing and scar hyperplasia, consistent with previous domestic and foreign research results.

Of course, it was important to pay proper attention to delicate surgical techniques. Careful management of the broken ureter end, degree of tissue dissociation, interruption time of the ureteral blood supply, and tension after ureteral replantation were considered during the operation. It remains cannot be accurately quantified and measured accurately.

There have been many previous studies trying to find increased risk factors for UIAS, but the exact cause is still unclear. Hoag et al. found in their research on risk factors for UIAS that diabetes and elevated levels increase the UIAS rate. It may be due to microvascular disease that the distal ureter becomes sensitive (9). In addition, age, body mass index, hemoglobin level, and ASA were not predictive factors for UIAS formation, which was consistent with our results.

Large et al. suggested that the running anastomosis and postoperative urinary tract infection might be related to UIAS. He believed that interrupted anastomosis had less effect on the blood supply at the anastomotic site, therefore the stricture rate was lower (10). In our study, there was no significant difference in the stricture rate between interrupted anastomosis and running anastomosis. Studies in our center currently suggest that running anastomoses have no significant effect on blood supply at the uretero-ileal anastomosis. Meanwhile, the influence of postoperative urinary tract infection on the stenosis rate was consistent with the study.

In previous reports on the prediction model of the incidence of radiotherapy and UIAS, it was identified that irradiated tissue has abnormal maturation of fibroblasts, leading to delayed healing, fibrosis, and scar formation in the distal ureter, as well as radiation-induced endarteritis that increased the risk of UIA ischemia. However, Katkoori et al. evaluated previous pelvic radiation for UIAS in 526 patients who had RC and UD between 1992 and 2008. They suggested that there was no significant difference between those with previous pelvic radiotherapy (pRT) and those without previous pRT (1.5% vs. 1.6%, P = 0.6) (3).

The conclusions on the influence of different surgical methods on the incidence of UIAS were different. Davis et al. found that there was no significant difference in the incidence of UIAS for Bricker and Wallace anastomoses (11). On the contrary, Kouba et al. found that Wallace anastomoses had a lower risk than Bricker anastomoses, but did not rule out a BMI effect on the results (12). In our study, there was no significant difference in the incidence of stenosis between Wallace and Bricker, meanwhile, and BMI had no significant effect on UIAS.

In the Mullins study, for the 192 patients with radical resection of the bladder and the UD retrospective study, it was found that in patients with the Wallace and Bricker operation method, stenting of the UIAS for the postoperative stricture rate has no obvious influence but is decreased after the incidence of intestinal obstruction (13). In our model, all patients underwent postoperative ureteral stent implantation to help control the formation of UIAS by transforming risk factors, and the effect of UIAS could not be determined. Current clinical experience shows that ureteral stents do not increase the rate of strictures. In the current study, the left UIAS was more marked than the right UIAS, as the left ureter passed beneath the sigmoid mesentery, increasing mobilization and tunneling under the sigmoid colon. However, in our model, although the left stricture rate was higher than the right stricture, there was no statistically significant difference (p = 0.54).

Anderson et al. in their study found that there was no significant difference in the rate of stricture between the robot-assisted and open groups (12.6% vs. 8.5%, p = 0.21) (14). In our institution, all undergoing radical cystectomy and ileal orthotopic bladder substitution patients adopt laparoscopic and robot assisted surgery and no significant difference in robot-assisted group and laparoscopic group.

Richards et al. found the median time to stricture formation on the right and left ureters to be 235 and 232 days, respectively. Besides, the length of the distal ureter resected did not significantly influence the stricture rate. The reason for the benign UIAS following was multifactorial (7).

There were several possible explanations for the finding of extracorporeal neobladder anastomoses as a risk factor for modified UIAS. The free length of the ureter in the external bladder anastomosis is 5–10 cm longer than the free length of the ureter in the internal bladder anastomosis, and the blood supply of the ureter is more affected. Besides, experienced surgeons choose a larger proportion of the intracorporeal neobladder anastomoses, better protection of the ureteral blood supply, more delicate tissue processing, more detailed surgical skills, less probability of uretero-ileum anastomosis leakage, and therefore a lower rate of stricture.

Nevertheless, this study had some limitations that are important to mention. For the retrospective studies and due to many confounding factors of collected data, retrospective studies often have substantial advantages to biomarker evaluation. The cases included in this study are all from the Zhejiang Provincial People's Hospital, which may be biased in the establishment of risk prediction models. Future research can increase the sample size or multi-center data to reduce the bias in experiment inclusion and improve the accuracy of risk assessment model predictions. Since some patients may have stricture but have no symptoms, it is still unknown whether the rate of stricture was not detected in time.

## Conclusions

Benign UIAS after ileal UD had a multifactorial etiology. In our series, intraoperative or postoperative blood transfusion, postoperative urinary tract infection, and extracorporeal neobladder anastomoses increased the risk of UIAS after radical cystectomy and ileal ureteral diversion. In addition, reducing the dissociation of the distal end of the ureter and protecting the blood supply of the ureter are essential to reducing the rate of stricture.

## Data availability statement

The original contributions presented in the study are included in the article/supplementary material. Further inquiries can be directed to the corresponding authors.

## Author contributions

ZL, BZ, and YH: Data collection, Data analysis, Manuscript writing. HL, XQ, SW, HW, and PZ: Data analysis. QW, LS, XH, YM, and FL: Project development. JC and DZ: Critical revision of manuscript. All authors contributed to the article and approved the submitted version.

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## Conflict of interest

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

## Publisher's note



## References

1. Wong M, Fung F, Leung C, Cheung W, Goggins WB, Ng CF. The global epidemiology of bladder cancer: A joinpoint regression analysis of its incidence and mortality trends and projection. *Sci Rep* (2018) 8:1129. doi: 10.1038/s41598-018-19199-z

2. Berdik C. Unlocking bladder cancer. *Nature* (2017) 551:S34–5. doi: 10.1038/551S34a

3. Katkoori D, Samavedi S, Adiyat KT, Soloway MS, Manoharan M. Is the incidence of uretero-intestinal anastomotic stricture increased in patients undergoing radical cystectomy with previous pelvic radiation? *Bju Int* (2010) 105:795–8. doi: 10.1111/j.1464-410X.2009.08835.x

4. Wishahi MM, Elganzoury H, Elkhouly A, Mehena A. Dipping technique for ureteroileal anastomosis in orthotopic ileal neobladder: 20-year experience in 670 patients-no stenosis with preservation of the upper tract. *ISRN Urol* (2013) 2013:725286. doi: 10.1155/2013/725286

5. De Sutter T, Akand M, Albersen M, Everaerts W, Van Cleynenbreugel B, De Ridder D, et al. The n-shaped orthotopic ileal neobladder: Functional outcomes and complication rates in 119 patients. *Springerplus* (2016) 5:646. doi: 10.1186/s40064-016-2287-1

6. Westerman ME, Parker WP, Viers BR, Rivera ME, Karnes RJ, Frank I, et al. Malignant ureteroenteric anastomotic stricture following radical cystectomy with urinary diversion: Patterns, risk factors, and outcomes. *Urol Oncol* (2016) 34:481–5. doi: 10.1016/j.urolonc.2016.06.008

7. Richards KA, Cohn JA, Large MC, Bales GT, Smith ND, Steinberg GD. The effect of length of ureteral resection on benign ureterointestinal stricture rate in ileal conduit or ileal neobladder urinary diversion following radical cystectomy. *Urol Oncol* (2015) 33:61–5. doi: 10.1016/j.urolonc.2014.05.015

8. Hosseini A, Dey L, Laurin O, Adding C, Hoijer J, Ebbing J, et al. Ureteric stricture rates and management after robot-assisted radical cystectomy: A single-centre observational study. *Scand J Urol* (2018) 52:244–8. doi: 10.1080/21681805.2018.1465462

9. Hoag N, Papa N, Beharry BK, Lawrentschuk N, Chiu D, Sengupta S, et al. Diabetes and elevated urea level predict for uretero-ileal stricture after radical cystectomy and ileal conduit formation. *Can Urol Assoc J* (2017) 11:E88–92. doi: 10.5489/cuaj.3848

10. Large MC, Cohn JA, Kiriluk KJ, Dangle P, Richards KA, Smith ND, et al. The impact of running versus interrupted anastomosis on ureterointestinal stricture rate after radical cystectomy. *J Urol* (2013) 190:923–7. doi: 10.1016/j.juro.2013.02.091

11. Davis NF, Burke JP, McDermott T, Flynn R, Manecksha RP, Thornhill JA. Bricker versus wallace anastomosis: A meta-analysis of ureteroenteric stricture rates after ileal conduit urinary diversion. *Can Urol Assoc J* (2015) 9:E284–90. doi: 10.5489/cuaj.2692

12. Kouba E, Sands M, Lentz A, Wallen E, Pruthi RS. A comparison of the bricker versus wallace ureteroileal anastomosis in patients undergoing urinary diversion for bladder cancer. *J Urol* (2007) 178:945–9. doi: 10.1016/j.juro.2007.05.030

13. Mullins JK, Guzzo TJ, Ball MW, Pierorazio PM, Eifler J, Jarrett TW, et al. Ureteral stents placed at the time of urinary diversion decreases postoperative morbidity. *Urol Int* (2012) 88:66–70. doi: 10.1159/000335212

14. Anderson CB, Morgan TM, Kappa S, Moore D, Clark PE, Davis R, et al. Ureteroenteric anastomotic strictures after radical cystectomy-does operative approach matter? *J Urol* (2013) 189:541–7. doi: 10.1016/j.juro.2012.09.034

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# Patients' related sexual outcomes in colorectal surgery

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**Background:** Patients undergoing colorectal surgery (CRS) have an increased risk of developing sexual disorders, attributed to different mechanisms. In this context, sexual function (SF) assessment of patients before and after surgery is essential: to identify risk factors for sexual disorders as well as to minimize their impact on overall quality of life (QoL), allowing them a satisfying relationship and sexual life.

**Material and methods:** Patients over 18 years of age who underwent a CRS in the University Hospital of Geneva, Switzerland, between June 2014 and February 2016 were included. Our main objective was to compare and analyze the evolution of SF, QoL, and marital satisfaction (MS) before and after CRS. Specific and standardized tests were used.

**Results:** A cohort of 72 patients with a median age of 58.73 was analyzed. The majority of CRS was elective (91.5%). A percentage of 52.8% of patients underwent surgery for oncological reasons. There was no statistical difference in SF, sexual QoL, and MS before and after elective or emergency CRS for men. Interestingly, a significant decrease in women's SF (FSFI) as well as their satisfaction within their couple (Locke–Wallace) until 12 months after surgery was found ($p = 0.021$). However, they showed a steady SF (GRISS) within their couple until 12 months after surgery.

**Conclusion:** Regarding knowledge about difficulties to talk about this intimate topic and gender differences, this general overview raises the question of the necessity to introduce in a long-course follow-up different methods of sexual health assessment with specific stakeholders.

KEYWORDS

**sexual function, marital satisfaction, colorectal surgery, assessment, patient related outcome**

**Abbreviations:** IIEF, International Index of Erectile Function; FSFI, Female Sexual Function Index; GRISS, Golombok, Rust Inventory of Sexual Satisfaction; Locke–Wallace, Locke–Wallace relationship adjustment test.

# Introduction

Colic and rectal resections are common procedures performed daily within a department of general surgery. It encompasses many benign and malign pathologies. The majority of publications linked to colorectal surgery aim at highlighting different kinds of outcome such as mortality, morbidity, and oncological/disease results.

However, with the improvement in the management of colorectal pathologies due to minimally invasive techniques (laparoscopy, robot-assisted surgery) (1), the adjunction of therapies like chemotherapy and/or radiotherapy in case of cancer (2), but also with the increase of inflammatory bowel disease (IBD) diagnosis especially among young patients (3, 4), it is necessary to assess specific outcomes after colorectal surgery. Thus, functional results and potential complications within the domain of sexuality have to be evaluated. Sexual function is one of the aspects of quality of life that may be disrupted after surgery. In case of colorectal surgery, sexual disorders appear to be multifactorial. During dissection, the superior and/or inferior hypogastric plexus may be damaged and linked to major sexual disorders like erectile dysfunction, problems of ejaculation, decrease of libido or lubrification, and dyspareunia (5). Psychological stress and body image modifications due to the surgery also imply sexuality alterations (6). Finally, the type of colorectal pathology with its dissemination/extension in the pelvis (colorectal cancers, inflammatory bowel diseases) and also its specific medical treatments can modulate sexual functions (7).

The aim of this study is to assess the impact of colorectal surgery on both sexual function and quality of life of patients and their partners.

# Materials and methods

## Data source

This monocentric prospective study focused on patients who underwent a colorectal surgery between June 2014 and February 2016 at Geneva University Hospitals, Switzerland, and was approved by the Health Research Ethics Board at the University of Geneva (CER 14-111).

## Patient population

Inclusion criteria were heterosexual patients in a stable relationship understanding French and having benefited from elective or emergency colorectal surgery in Geneva University Hospital for colorectal cancer or diverticular or bowel inflammatory diseases. No patient was included in the database twice.

Pregnant patients, patients under 18 years of age, those who have fulfilled only one questionnaire, those who died during the study, patients without sexual activity, patients with tumoral progression during the follow-up or having left the study, and homosexual patients were excluded.

## Methods

This study compared sexual function and marital satisfaction before and after colorectal surgery in both men and women.

All participating subjects provided written informed consent.

## Questionnaires

Validated questionnaires were given to patients waiting for elective or emergency colorectal surgery before (before surgery or during the hospital stay according to the degree of emergency) and after (at 3, 6, and 12 months follow-up) surgery. They were filled out without help.

For the assessment of the sexual function, gender-specific questionnaires were used: the International Index of Erectile Function for men [IIEF] and the Female Sexual Function Index for women [FSFI].

For the evaluation of the quality of life, the Locke–Wallace relationship adjustment test [marital satisfaction] and the GRISS [quality of sexual life] were used.

The IIEF is a 15-item questionnaire, assessing all dimensions of male sexual function: erectile function, orgasmic function, sexual desire, intercourse satisfaction, and overall satisfaction (8). Each item is scored on a five-point scale, and the overall score (OS) (minimum: 5 to maximum: 75 points) is obtained by adding each item score. Erectile dysfunction (ED) is classified as severe ED (OS between 1 and 10), mild to moderate ED (11–25), and no ED (≥26).

The FSFI questionnaire is a 19-item questionnaire, assessing all aspects of female sexual function: desire, arousal, lubrication, orgasm, satisfaction, and pain (9). Each item is scored on a six-point scale with an OS (minimum of 2 and maximum of 36 points) obtained by combining each item score. An OS lower than 23 defines a poor sexual function, an OS between 23 and 29 means a good sexual function, and an OS greater than 29 corresponds to a very good sexual function. The overall score is lower than 26 in the presence of one or more dysfunctions in specific areas.

The Locke–Wallace Marital Adjustment Test is a 15-item test, assessing the level of couple satisfaction by underlining the extension of agreement/disagreement between partners. Each item is scored from 0 to 35. OS (a minimum 2 and a maximum of 158 points) is obtained by adding each item score. The severity of issues encountered by the partners can be classified as serious (OS <80), difficulties (OS 80–100), and no problems (OS >100) (10).

The GRISS is a 28-item questionnaire assessing the existence and severity of sexual problems within the couple and for each partner. There is a version for each gender. Various aspects of the relationship are explored: communication, non-genital physical contact, dissatisfaction, avoidance of sexual intercourses, frequency of sexual activity, and impotence and premature ejaculation for men and anorgasmia and vaginismus for women. The obtained overall score measures the sexual dysfunction: the higher the score, the greater the sexual dysfunction. The score ranges from 0 to 10, and values higher than 5 indicate sexual dysfunction.

## Outcomes and covariates definition

The main outcome was the evolution of both sexual function and marital satisfaction of patients after colorectal surgery.

## Statistical analysis

Results are expressed as medians with interquartile range (IQR) for quantitative variables and qualitative variables.

Comparisons before and after surgery were done with ANOVA test (analysis of variance).

In order to bring out a significant change concerning the different tests before and after surgery, inclusion of a minimum of 20 patients in each group was necessary. Around 200 patients are operated each year for a colorectal pathology in the department, and considering a minimum attendance, a satisfactory statistical power could be achieved in 2 years.

# Results

Questionnaires were proposed to 103 patients. Of these patients, 31 did not meet the inclusion criteria. After the exclusion process, 72 patients were included (Figure 1).

Among the 72 patients, 56 (77.8%) were men and 16 (22.2%) were women. Patients' characteristics are described in Table 1 with a median age of 58.73 years (50.37–68.54). The median body mass index (BMI) was 25.60 kg/m² (23.58–28.40).

Various comorbidities were present within the cohort: active smoking (42.9%), cardiovascular background (31.0%), regular alcohol consumption (26.8%), dyslipidemia (18.3%), immune insufficiency (11.6%, due to immunosuppressive therapy such corticosteroids), psychiatric disorders (11.3%), and diabetes (4.2%).

Most surgical interventions (91.5%) were elective procedures. In 52.8% of cases, surgery was performed for oncological reasons and in 30.6% for benign non-inflammatory bowel diseases. The disease had double localization (small and large intestine) in 8.4% of cases whereas it was only localized in the colon in 19.4%, in the rectum in 23.6%, or in the sigmoid in 48.6%. There were 17 of 38 oncological patients (44.7%) who received a neoadjuvant treatment.

## Preoperative setting

At the time of surgery, women had a mean FSFI overall score of 28 indicating a good sexual function, a mean overall score for the Locke–Wallace Marital Adjustment Test of 118 meaning a good agreement within the couple, and a mean GRISS score of 61 (Tables 2, 3).

For men, the preoperative mean IIEF score was 49 corresponding to no erectile dysfunction, the mean overall score for the Locke–Wallace Martial Adjustment Test was 119 showing no disagreement between partners, and the mean GRISS score was 62 (Tables 2, 4).

## Postoperative setting and evolution

After surgery, women indicated that their own sexual function (FSFI) slightly decreased until 12 months (Figure 2) as well as their satisfaction within their couple (Locke–Wallace Marital Adjustment Test) ($p = 0.021$ between LWAT scores before surgery and 6 months after surgery) (Figure 2**).** However, they showed a steady sexual function (GRISS) within their couple until 12 months after surgery (Figure 2).

Regarding men, they assessed their own sexual function as quite stable (IIEF) until 12 months after surgery as well as their satisfaction within their couple (Locke–Wallace) and their sexual function within their couple (GRISS) (Figure 3).

# Discussion

One of the interests of this study is to highlight sexuality, a subject too often taboo for caregivers but nevertheless essential for the quality of life of their patients. Thanks to this research, we have a detailed view of the sexual health of the patients as well as their development within their couple before surgery. Interestingly, we find that patients waiting for elective surgery have an overall satisfactory sexuality and relationship.

Moreover, this study demonstrates that colorectal surgery regardless of indication (including inflammatory disease and oncological pathology) and location of the intestinal resection

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TABLE 1 Characteristics of the population included.

| | Female (N = 16) | Male (N = 56) | Total (N = 72) |
|---|---|---|---|
| Age | | | |
| Median (Q1, Q3) | 56.59 (48.15, 68.67) | 58.73 (50.65, 68.48) | 58.73 (50.37, 68.54) |
| Min–max | 36.98–73.37 | 25.69–83.89 | 25.69–83.89 |
| Indication | | | |
| Adenoma (low-grade dysplasia) | 0 (0.0%) | 1 (1.8%) | 1 (1.4%) |
| Inflammatory bowel disease | 1 (6.2%) | 9 (16.1%) | 10 (13.9%) |
| Cancer | 9 (56.2%) | 29 (51.8%) | 38 (52.8%) |
| Non-inflammatory bowel disease | 6 (37.5%) | 16 (28.6%) | 22 (30.6%) |
| Polyp (high-grade dysplasia) | 0 (0.0%) | 1 (1.8%) | 1 (1.4%) |
| Localization | | | |
| Unique | | | |
| Right colon | 1 (6.2%) | 7 (12.5%) | 8 (11.1%) |
| Transverse colon | 0 (0.0%) | 1 (1.8%) | 1 (1.4%) |
| Left colon | 2 (12.5%) | 3 (5.4%) | 5 (6.9%) |
| Sigmoid | 8 (50.0%) | 27 (48.2%) | 35 (48.6%) |
| Rectum | 4 (25.0%) | 13 (23.2%) | 17 (23.6%) |
| Double | | | |
| Colon and rectum | 0 (0.0%) | 1 (1.8%) | 1 (1.4%) |
| Ileum and cecum | 1 (6.2%) | 3 (5.4%) | 4 (5.6%) |
| Sigmoid and appendix | 0 (0.0%) | 1 (1.8%) | 1 (1.4%) |
| Neoadjuvant radiochemotherapy | | | |
| No | 8 (66.7%) | 38 (74.5%) | 46 (73.0%) |
| Yes | 4 (33.3%) | 13 (25.5%) | 17 (27.0%) |
| Emergency | | | |
| No | 16 (100.0%) | 49 (89.1%) | 65 (91.5%) |
| Yes | 0 (0.0%) | 6 (10.9%) | 6 (8.5%) |
| Diabetes | | | |
| No | 16 (100.0%) | 52 (94.5%) | 68 (95.8%) |
| Yes | 0 (0.0%) | 3 (5.5%) | 3 (4.2%) |
| Smoking | | | |
| No | 9 (56.2%) | 31 (57.4%) | 40 (57.1%) |
| Yes | 7 (43.8%) | 23 (42.6%) | 30 (42.9%) |
| Immunodepression | | | |
| No | 14 (87.5%) | 47 (88.7%) | 61 (88.4%) |
| Yes | 2 (12.5%) | 6 (11.3%) | 8 (11.6%) |
| BMI | | | |
| Median (Q1, Q3) | 25.85 (22.67, 28.63) | 25.45 (23.60, 28.40) | 25.60 (23.58, 28.40) |
| Min–max | 19.00–36.10 | 19.80–33.50 | 19.00–36.10 |
| Psychiatric_disorder | | | |
| No | 13 (81.2%) | 50 (90.9%) | 63 (88.7%) |
| Yes | 3 (18.8%) | 5 (9.1%) | 8 (11.3%) |
| Dyslipidemia | | | |
| No | 12 (75.0%) | 46 (83.6%) | 58 (81.7%) |
| Yes | 4 (25.0%) | 9 (16.4%) | 13 (18.3%) |
| Alcohol | | | |
| No | 15 (93.8%) | 37 (67.3%) | 52 (73.2%) |
| Yes | 1 (6.2%) | 18 (32.7%) | 19 (26.8%) |
| Cardiovascular pathology | | | |
| No | 13 (81.2%) | 36 (65.5%) | 49 (69.0%) |
| Yes | 3 (18.8%) | 19 (34.5%) | 22 (31.0%) |

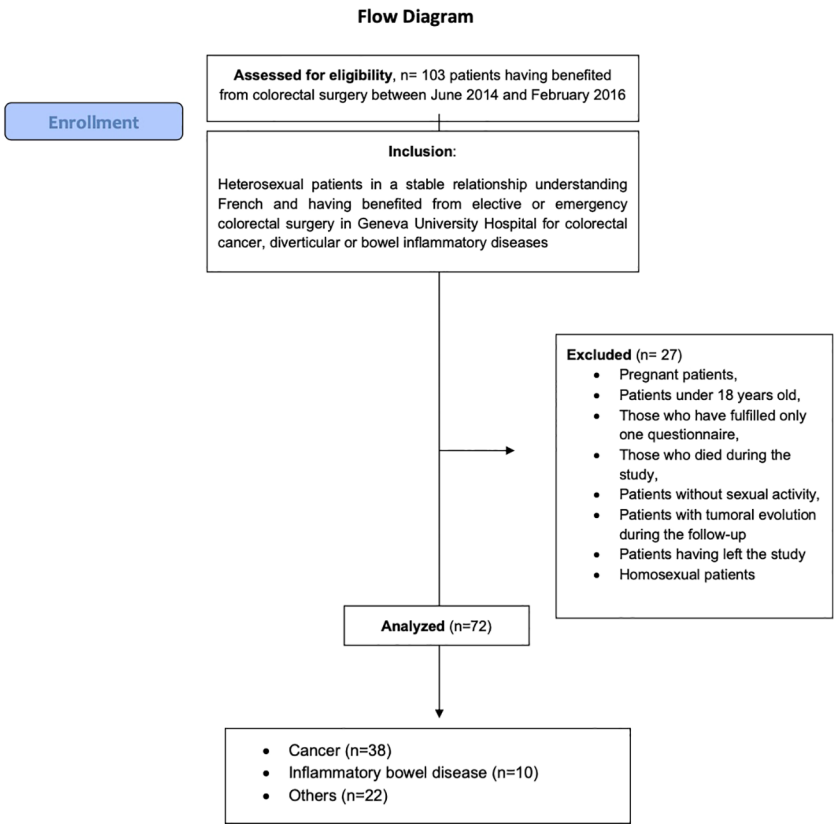



FIGURE 1
Flow diagram.

(right colon, transverse colon, sigmoid, rectum, or both) is not linked to modifications of sexual functions, sexual quality of life, or marital satisfaction.

After reviewing the literature, most studies in the field of colorectal surgery analyze mortality and morbidity and aim to identify their risk factors. However, few of them are interested in functional complications of such procedures like sexual dysfunctions. These are focused on oncological surgery and especially in the subgroup of men (11) and the rectal location (12–14).

Indeed, in colorectal surgery, the surgical procedures of the rectum are more concerned about sexual dysfunctions as pelvic localization means dissection along the superior and/or inferior hypogastric plexus contributing to the innervation of

TABLE 2 Overall results for marital satisfaction and sexual quality of life.

| | Preoperative period (N = 72) | 3 months after surgery (N = 72) | 6 months after surgery (N = 72) | 12 months after surgery (N = 72) | Total (N = 288) |
|---|---|---|---|---|---|
| **GRISS total** | | | | | |
| Median (Q1, Q3) | 64.0 (58.5, 66.5) | 62.0 (59.5, 66.0) | 63.0 (59.7, 67.0) | 61.0 (57.5, 65.0) | 63.0 (58.0, 66.0) |
| Min–max | 45.0–93.0 | 44.0–85.0 | 53.0–73.0 | 53.0–76.0 | 44.0–93.0 |
| **Locke–Wallace** | | | | | |
| Median (Q1, Q3) | 124.5 (103.2, 139.0) | 118.0 (100.5, 135.7) | 120.5 (105.2, 132.7) | 121.5 (97.0, 131.7) | 121.5 (101.2, 136.0) |
| Min–max | 12.0–156.0 | 14.0–157.0 | 34.0–148.0 | 37.0–151.0 | 12.0–157.0 |

TABLE 3 Summary outcomes for women.

| | preop (N = 16) | 3 months (N = 16) | 6 months (N = 16) | 12 months (N = 16) | Total (N = 64) |
|---|---|---|---|---|---|
| GRISS_total | | | | | |
| Mean (SD) | 65.14 (9.27) | 62.50 (5.80) | 67.82 (4.38) | 65.36 (6.12) | 65.26 (6.89) |
| Median (Q1, Q3) | 63.50 (61.00, 67.00) | 63.00 (60.00, 66.25) | 68.00 (63.50, 71.50) | 63.00 (61.50, 69.50) | 64.00 (61.25, 69.75) |
| Min–max | 53.00–93.00 | 50.00–70.00 | 62.00–73.00 | 57.00–76.00 | 50.00–93.00 |
| Locke–Wallace | | | | | |
| Mean (SD) | 119.53 (24.20) | 124.90 (18.16) | 104.70 (33.96) | 107.82 (30.45) | 114.67 (27.38) |
| Median (Q1, Q3) | 126.00 (104.00, 137.50) | 128.50 (113.75, 135.75) | 119.50 (83.00, 126.50) | 121.00 (84.50, 130.50) | 124.50 (102.25, 134.75) |
| Min–max | 63.00–152.00 | 92.00–152.00 | 34.00–140.00 | 52.00–141.00 | 34.00–152.00 |
| IFSF_total | | | | | |
| Mean (SD) | 28.09 (5.40) | 24.97 (9.51) | 19.18 (12.52) | 22.68 (8.52) | 24.15 (9.32) |
| Median (Q1, Q3) | 30.00 (26.25, 31.75) | 30.00 (22.70, 30.90) | 20.60 (7.45, 30.60) | 23.25 (19.23, 28.52) | 27.30 (19.80, 31.17) |
| Min–max | 14.10–35.40 | 2.90–33.00 | 1.90–34.80 | 2.30–35.00 | 1.90–35.40 |

genital organs. Moreover, men are more concerned by colorectal cancer: around 1 million of men versus 846,000 women in 2020 worldwide (15) despite a trend to women in some countries (16).

Following this statement, at first sight, one limit of our work is to have encompassed both localization of the colon and rectum. However, it is important here to stress out that colon surgery in the right or transverse colon, which are not localized into the pelvis, and regardless of indication for surgery, may also lead to sexual dysfunctions. Indeed, sexual function is linked to several factors. Among them, we can point out psychological factors (stress, apparition of a disease, modifications of the body image with presence of potential stoma bag, issues within relationships, patients' social situation, etc.) and physical issues due to treatments (medications, chemotherapy, biotherapy, radiotherapy, surgery). Consequently, not only rectal surgery with its specific localization but also colonic surgery should be given special attention regarding sexual dysfunctions through a biopsychomedical perspective.

In our study, we used homogenous and validated tests concerning sexual domains.

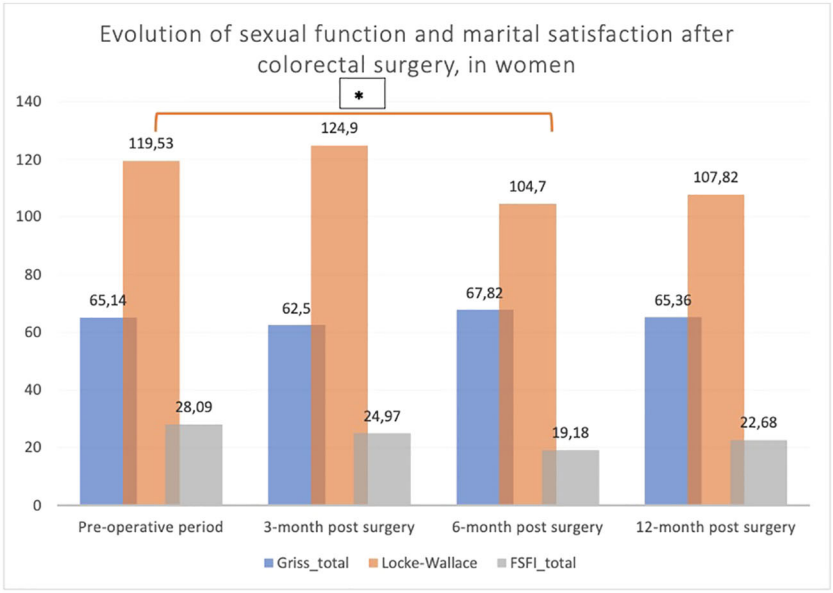
Our results regarding sexual functions are in accordance with those of Traa et al. (17), where no changes were found before and after colorectal cancer surgery.

Interestingly, we found a significant decrease in women's sexual function (FSFI) as well as their satisfaction within their couple (Locke–Wallace) until 12 months after surgery. In contrast, they showed a steady sexual function (GRISS) within their couple until 12 months after surgery.

Regarding men, the evolution over time for both their own sexual function and within the couple but also their satisfaction within the couple seems stable.

TABLE 4 Summary outcomes for men.

| | preop (N = 56) | 3 months (N = 56) | 6 months (N = 56) | 12 months (N = 56) | Total (N = 224) |
|---|---|---|---|---|---|
| GRISS total | | | | | |
| Mean (SD) | 61.67 (6.37) | 61.24 (8.11) | 61.31 (4.69) | 60.53 (4.27) | 61.23 (6.08) |
| Median (Q1, Q3) | 64.00 (56.00, 66.00) | 62.00 (58.00, 66.00) | 61.00 (57.00, 65.00) | 60.00 (57.00, 63.50) | 62.00 (57.00, 65.00) |
| Min–max | 45.00–73.00 | 44.00–85.00 | 53.00–70.00 | 53.00–70.00 | 44.00–85.00 |
| Locke–Wallace | | | | | |
| Mean (SD) | 118.96 (26.54) | 111.75 (31.35) | 119.22 (20.80) | 113.95 (27.61) | 116.09 (26.89) |
| Median (Q1, Q3) | 122.00 (103.50, 141.50) | 111.00 (98.50, 132.50) | 120.50 (109.75, 137.50) | 122.00 (98.00, 131.00) | 120.00 (100.75, 137.00) |
| Min–max | 12.00–156.00 | 14.00–157.00 | 80.00–148.00 | 37.00–151.00 | 12.00–157.00 |
| IIEF total | | | | | |
| Mean (SD) | 48.84 (19.84) | 46.59 (18.55) | 49.32 (18.70) | 48.49 (21.50) | 48.26 (19.57) |
| Median (Q1, Q3) | 51.50 (41.50, 65.00) | 48.00 (35.00, 59.75) | 56.00 (38.00, 62.25) | 53.00 (35.00, 67.00) | 52.00 (36.50, 65.00) |
| Min–max | 7.00–74.00 | 5.00–71.00 | 5.00–74.00 | 5.00–75.00 | 5.00–75.00 |

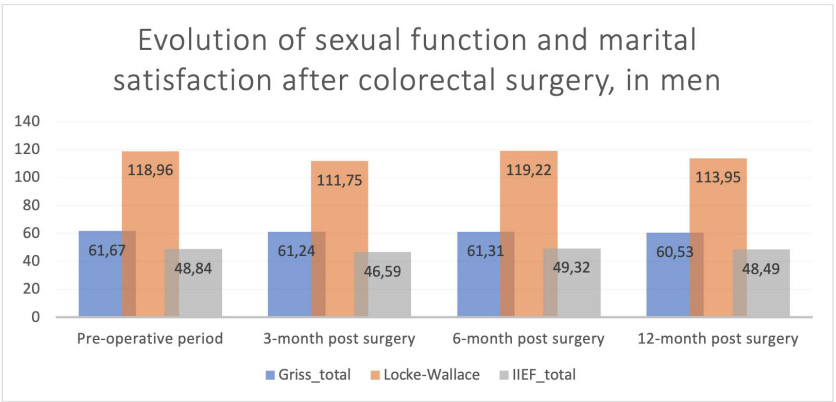



FSFI score, Locke-Wallace Marital Adjustment Test and GRISS score are expressed in mean (SD)

*p= 0.021 between Locke-Wallace – Women before surgery and Locke-Wallace – Women 6 months after surgery

FIGURE 2
Evolution of sexual function and marital satisfaction after colorectal surgery, in women.

Thus, these results and differences seem to point out the necessity to conduct a deeper analysis with different methodologies to assess the accuracy and veracity of these answers. Indeed, it is well-known that there are gender-related differences toward sexuality function and sexual health (18). Moreover, sexuality is a topic of privacy and in some ways may be difficult to speak about, needing trust and confidentiality (19). Even in the healthcare system, the subject brings caution and




IIEF score, Locke-Wallace Marital Adjustment Test and GRISS score are expressed in mean (SD)

FIGURE 3
Evolution of sexual function and marital satisfaction after colorectal surgery, in men.

sometimes has not even been considered in the discussion or assessed before surgery between the surgeon and the patient according to a recent survey (20).

This study did not have the ambition to focus and assess specific sexual dysfunctions or sexual unwell-being after specific pathologies or subgroup of women or men. It is more an overview about the topic of sexual function and health of patients undergoing colorectal surgery.

### Strengths

The major interest of our study, contrary to those of the current literature, is to analyze sexual dysfunctions after colorectal surgery whatever indication, localization, or gender. Contrary to other studies, our analysis was based on validated questionnaires.

### Limitations

The main limit of our work is the presence of a small number of persons especially in the subgroup of women where the number of 20 that was expected was not reached. This can be explained by the need of time for patients to fill out the questionnaires because they are detailed. Thus, some patients had the tests without having completed them, whereas others were total non-responders. Moreover, a longer follow-up would have been more suitable. Other limits can be pointed out and will have to be taken into account for the next research. More recent data after 2016 have to be studied and compared with these ones; all the colorectal procedures have been included in our work, which can be a reason for bias as the number of rectal cases is less and it is well documented that sexual function is mostly affected by rectal dissection. Nevertheless, it is important to highlight that there are many studies which deal with quality of life after rectal surgeries, but there are very limited studies which deal exclusively with sexual functions after such surgeries, making our study an important one. Our study covers an extremely wide group of patients (malignant–benign, colon–rectal resections, presence–absence of stoma). The fact that it was conducted in a small group precludes any subgroup analysis. Furthermore, we can underline that studies involving larger and different populations may be interesting and valuable: thus, in future studies, we will include homosexual and bisexual patients and those without an apparent "sexuality."

## Conclusion

Interestingly, in this monocentric study, colorectal surgery does not influence sexual function and sexual quality of life in both men and women whatever the indication until 1 year after surgery.

However, preservation of the sexual function as well as the marital satisfaction of colorectal patients should be of major concern for the involved caregivers, alongside outcomes like morbidity, mortality, or oncological results. Sexual disorders should be assessed, as other aspects of quality of life, before and after surgery to identify their occurrence and offer appropriate care. The use of standardized and validated questionnaires, if possible by involving the partner, ensures quality follow-up.

These results should be confirmed by larger multicentric studies.

## Data availability statement

The raw data supporting the conclusions of this article will be made available by the authors, without undue reservation.

## Ethics statement

This study was approved by the Health Research Ethics Board at the University of Geneva (CER 14-111). The patients/participants provided their written informed consent to participate in this study.

## Author contributions

EL, SdS, JK, NB, and FR conceived and designed the study. NC, EL, SdS, JK, and DG acquired the data. NC and EL analyzed the data. NC and EL interpreted the data. NC, EL, SdS, JK, IP, DG, NB, and FR contributed to the writing of the manuscript and to its critical revision. NC, EL, SdS, JK, IP, DG, NB, and FR approved the final version of the manuscript. All authors contributed to the article and approved the submitted version.

## Conflict of interest

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

## Publisher's note

# References


1. Bardakcioglu O. *Advanced techniques in minimally invasive and robotic colorectal surgery*. United States: Springer US (2015). Available at: https://www.springer.com/gp/book/9781489978318.

2. Falzone L, Salomone S, Libra M. Evolution of cancer pharmacological treatments at the turn of the third millennium. *Front Pharmacol* (2018) 9:1300/full. doi: 10.3389/fphar.2018.01300/full

3. Ng SC, Shi HY, Hamidi N, Underwood FE, Tang W, Benchimol EI, et al. Worldwide incidence and prevalence of inflammatory bowel disease in the 21st century: A systematic review of population-based studies. *Lancet Lond Engl* (2017) 390(10114):2769–78. doi: 10.1016/S0140-6736(17)32448-0

4. Mak WY, Zhao M, Ng SC, Burisch J. The epidemiology of inflammatory bowel disease: East meets west. *J Gastroenterol Hepatol* (2020) 35(3):380–9. doi: 10.1111/jgh.14872

5. Weledji E, Ngounou E. The-anatomical-basis-for-autonomic-dysfunction-in-pelvic-surgery. *Gen Surg* (2020) 14:4.

6. da Silva GM, Hull T, Roberts PL, Ruiz DE, Wexner SD, Weiss EG, et al. The effect of colorectal surgery in female sexual function, body image, self-esteem and general health: a prospective study. *Ann Surg* (2008) 248(2):266–72. doi: 10.1097/SLA.0b013e3181820cf4

7. Basson R, Schultz WW. Sexual sequelae of general medical disorders. *Lancet Lond Engl* (2007) 369(9559):409–24. doi: 10.1016/S0140-6736(07)60197-4

8. Rosen RC, Riley A, Wagner G, Osterloh IH, Kirkpatrick J, Mishra A. The international index of erectile function (IIEF): A multidimensional scale for assessment of erectile dysfunction. *Urology* (1997) 49(6):822–30. doi: 10.1016/S0090-4295(97)00238-0

9. Rosen R, Brown C, Heiman J, Leiblum S, Meston C, Shabsigh R, et al. The female sexual function index (FSFI): A multidimensional self-report instrument for the assessment of female sexual function. *J Sex Marital Ther* (2000) 26(2):191–208. doi: 10.1080/009262300278597

10. Locke HJ, Wallace KM. Short marital-adjustment and prediction tests: Their reliability and validity. *Marriage Fam Living* (1959) 21:251–5. doi: 10.2307/348022

11. Wang G, Wang Z, Jiang Z, Liu J, Zhao J, Li J. Male Urinary and sexual function after robotic pelvic autonomic nerve-preserving surgery for rectal cancer. *Int J Med Robot* (2017) 13(1):e1725. doi: 10.1002/rcs.1725

12. Adam J-P, Denost Q, Capdepont M, van Geluwe B, Rullier E. Prospective and longitudinal study of urogenital dysfunction after proctectomy for rectal cancer. *Dis Colon Rectum* (2016) 59(9):822–30. doi: 10.1097/DCR.0000000000000652

13. Kim HJ, Choi G-S, Park JS, Park SY, Yang CS, Lee HJ. The impact of robotic surgery on quality of life, urinary and sexual function following total mesorectal excision for rectal cancer: A propensity score-matched analysis with laparoscopic surgery. *Colorectal Dis Off J Assoc Coloproctol G B Irel* (2018) 20(5):O103–13. doi: 10.1111/codi.14051

14. Morelli L, Di Franco G, Guadagni S, Rossi L, Palmeri M, Furbetta N, et al. Robot-assisted total mesorectal excision for rectal cancer: Case-matched comparison of short-term surgical and functional outcomes between the da Vinci xi and Si. *Surg Endosc* (2018) 32(2):589–600. doi: 10.1007/s00464-017-5708-5

15. Sung H, Ferlay J, Siegel RL, Laversanne M, Soerjomataram I, Jemal A, et al. Global cancer statistics 2020: GLOBOCAN estimates of incidence and mortality worldwide for 36 cancers in 185 countries. *CA Cancer J Clin* (2021) 71(3):209–49. doi: 10.3322/caac.21660

16. Wong MCS, Huang J, Lok V, Wang J, Fung F, Ding H, et al. Differences in incidence and mortality trends of colorectal cancer worldwide based on sex, age, and anatomic location. *Clin Gastroenterol Hepatol Off Clin Pract J Am Gastroenterol Assoc* (2021) 19(5):955–966.e61. doi: 10.1016/j.cgh.2020.02.026

17. Traa MJ, Roukema JA, De Vries J, Rutten HJT, Langenhoff B, Jansen W, et al. Biopsychosocial predictors of sexual function and quality of sexual life: a study among patients with colorectal cancer. *Transl Androl Urol* (2015) 4(2):206–17. doi: 10.3978/j.issn.2223-4683.2015.03.01

18. Mollaioli D, Ciocca G, Limoncin E, Di Sante S, Gravina GL, Carosa E, et al. Lifestyles and sexuality in men and women: The gender perspective in sexual medicine. *Reprod Biol Endocrinol RBE* (2020) 18:10. doi: 10.1186/s12958-019-0557-9

19. Hinchliff S, Fileborn B, Alba B, Lyons A, Minichiello V, Barrett C, et al. Talking about sex with friends: perspectives of older adults from the sex, age & me study in Australia. *Cult Health Sex* (2021) 23(3):367–82. doi: 10.1080/13691058.2019.1710568

20. Dames NB, Squire SE, Devlin AB, Fish R, Bisset CN, Tozer P, et al. 'Let's talk about sex': A patient-led survey on sexual function after colorectal and pelvic floor surgery. *Colorectal Dis* (2021) 23(6):1524–51. doi: 10.1111/codi.15598

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# An effective tool for predicting survival in breast cancer patients with *de novo* lung metastasis: Nomograms constructed based on SEER

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**Background & objectives:** An effective tool for forecasting the survival of BCLM is lacking. This study aims to construct nomograms to predict overall survival (OS) and breast cancer-specific survival (BCSS) in breast cancer patients with *de novo* lung metastasis, and to help clinicians develop appropriate treatment regimens for breast cancer lung metastasis (BCLM) individuals.
**Methods:** We gathered clinical data of 2,537 patients with BCLM between 2010 and 2015 from the Surveillance, Epidemiology, and End Results (SEER) database. Cox regression analysis was employed to identify independent prognostic parameters for BCLM, which were integrated to establish nomograms by R software. The discriminative ability and predictive accuracy of the nomograms were assessed using the concordance index (C-index), receiver operating characteristic (ROC) curves, and calibration plots. Kaplan–Meier analyses were applied to evaluate the clinical utility of the risk stratification system and investigate the survival benefit of primary site surgery, chemotherapy, and radiotherapy for BCLM patients.
**Results:** Two nomograms shared common prognostic indicators including age, marital status, race, laterality, grade, AJCC T stage, subtype, bone metastasis, brain metastasis, liver metastasis, surgery, and chemotherapy. The results of the C-index, ROC curves, and calibration curves demonstrated that the nomograms exhibited an outstanding performance in predicting the prognosis of BCLM patients. Significant differences in the Kaplan–Meier curves of various risk groups corroborated the nomograms' excellent stratification. Primary site surgery and chemotherapy remarkably improved OS and BCSS of BCLM patients whether the patients were at low-risk or high-risk, but radiotherapy did not.
**Conclusions:** We successfully developed prognostic stratification nomograms to forecast prognosis in BCLM patients, which provide important information for indicating prognosis and facilitating individualized treatment regimens for BCLM patients.

KEYWORDS

breast cancer lung metastasis, nomogram, prognosis, SEER, survival

# Introduction

The incidence of breast cancer (BC) is highest among malignant tumors, and breast cancer is one of the leading causes of cancer-related death worldwide (1). When breast cancer patients are first diagnosed, approximately 5%–10% of them have distant metastasis (2). The lung is the second most common metastatic site in breast cancer patients (3). In a study encompassing 11,568 patients with metastatic breast cancer, 36.4% of patients had lung metastasis (4). Despite amelioration in diverse treatments, including radiotherapy, chemotherapy, or targeted therapy, the prognosis of breast cancer patients with lung metastasis remains poor with a median survival of 13 to 21 months (4, 5). In addition, a large proportion of breast cancer lung metastasis (BCLM) patients always suffer severe complications synchronously, leading to a high mortality rate in BCLM patients. The survival-related risk factors of BCLM have been reported (4), but an effective tool for forecasting the survival of BCLM is lacking.

Recently, nomograms have been extensively used in tumor prediction as a reliable predicted tool (6, 7). Thus, in this study, we exploited data from the Surveillance, Epidemiology, and End Results (SEER) database to identify independent prognostic factors associated with survival in BCLM patients, and developed nomograms to predict OS and BCSS in patients with BCLM. Besides, we built a risk stratification system based on the nomogram models and evaluated the benefit of different treatments in diverse stratified risk groups.

# Methods

## Data collection and study design

We used SEER*Stat 8.3.9 to acquire the data of adult patients who were primarily diagnosed with breast cancer lung metastasis between 2010 and 2015 ($n = 4{,}834$). Patient demographic characteristics (sex, age, marital status, and race), disease characteristics (site, laterality, grade, American Joint Committee on Cancer (AJCC) T stage, AJCC N stage, molecular type, and distant metastatic sites), treatment modalities (surgery, chemotherapy, and radiotherapy) and survival status (survival time, vital status and cause of death) were included in our study. The selection process of detailed inclusion and exclusion criteria is displayed in **Figure 1**. Eventually, 2,537 eligible patients were extracted for further study. There was no need for formal consent in this type of retrospective study.

## Statistical analysis

We randomly allocated eligible patients into training and validation cohorts at a ratio of 7:3. According to the cause of death classification in the SEER database, the time from the date of diagnosis to death from any cause was defined as overall survival (OS), and the time from the date of diagnosis to the date of death from breast cancer was defined as breast cancer-specific survival (BCSS).

The characteristics of the training cohort and the validation cohort were compared using the chi-squared test. Univariate and multivariate Cox analyses were utilized to identify independent risk factors for prognosis. All of the identified independent risk factors were employed to construct nomograms for estimating 1-, 2-, and 3-year OS and BCSS. The discriminative ability of the nomograms was assessed using the C-index and ROC curves. The predictive capacity of the nomograms was tested by calibration plots, which can estimate the predicted and observed survival probability. Based on the aggregate score of the clinicopathological baseline data in the nomograms, breast cancer patients with lung metastasis were divided into low-risk and high-risk groups. Kaplan–Meier survival analyses were applied to assess the discriminatory power of the risk stratification system and investigate the survival benefit of primary site surgery, chemotherapy, and radiotherapy for BCLM patients in different risk groups.

All of these analyses were executed using packages (including caret, rms, foreign, survival, and survivalROC) in R software (version 4.0.4; http://www.r-project.org). A two-sided $P < 0.05$ was considered statistically significant.

# Results

## Patient characteristics

Through rigorous selection, as shown in **Figure 1**, a total of 2,537 breast cancer patients with initial lung metastasis were included for analysis (1,777 patients in the training set and 760 patients in the validation set). Patients of the entire cohort were found to have a median survival time of 25 months (95% CI: 24–27), and have 0.695 (95% CI: 0.677–0.713), 0.509 (95% CI: 0.490–0.529), 0.388 (95% CI 0.369–0.407) of 1-, 2-, 3- year survival rates respectively. Demographics and clinicopathologic characteristics of BCLM patients were displayed in **Table 1**. In the training cohort, most of the patients were female (98.4%, 1748) and white (72.8%, 1294), and the age of patients was mainly distributed among middle-aged and senior people (40–59 years old: 38.4%, 682; 60–79 years old: 44.1%, 784). BCLM patients with higher grades and higher T stages accounted for a higher

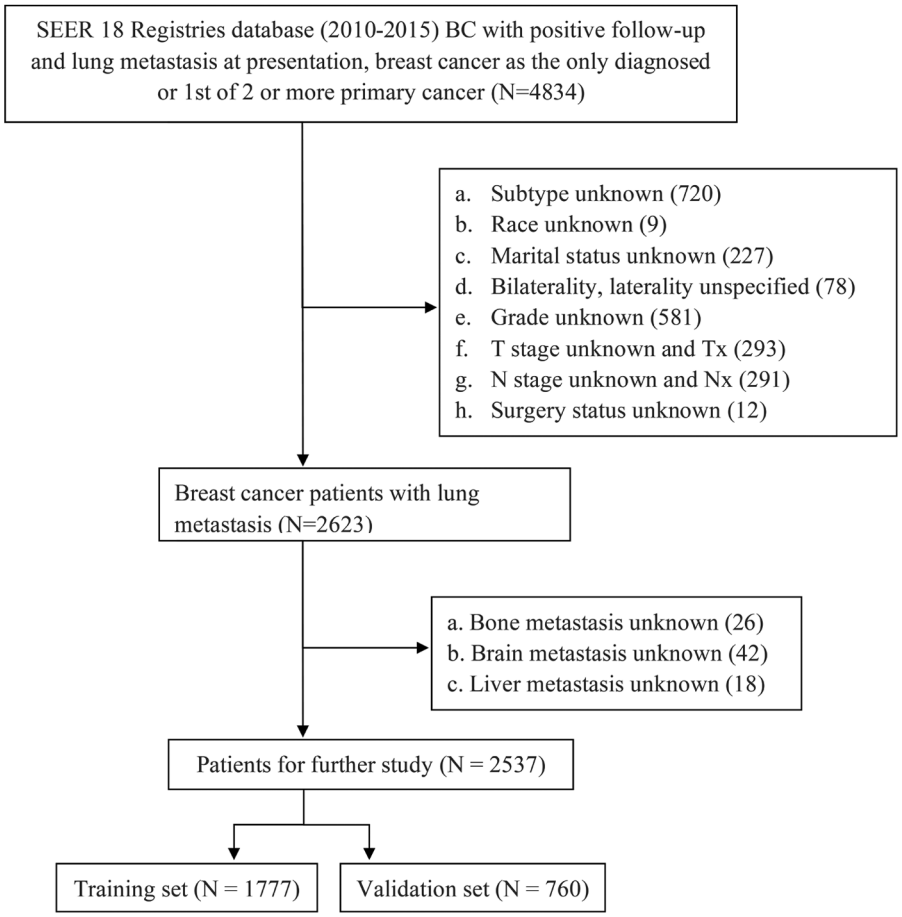


FIGURE 1
The flowchart of patients selected in the present study.

proportion. Moreover, more than half of BCLM patients were HR+/HER2-. Furthermore, the proportion of chemotherapy-received patients was almost two times of the surgery- or radiotherapy-received, 65.5%, 32.6%, and 30.0% in the training cohort, respectively. In addition, the incidence of bone metastasis in BCLM patients was the highest (53.0%), followed by liver metastasis (28.1%).

## Univariate and multivariate cox regression analysis

The results generated by univariate Cox analysis are listed in **Supplementary Table S1**. We identified twelve variables including age, marital status, race laterality, grade, AJCC T stage, subtype, bone metastasis, brain metastasis, liver metastasis, surgery, and chemotherapy, that were statistically associated with the OS and BCSS of BCLM patients. These twelve variables were included in multivariate analysis, and the results suggested that all of the twelve variables were confirmed as final prognostic factors for OS and BCSS (**Table 2**).

## Construction and validation of nomograms

The twelve final prognostic factors were then used in the nomogram establishment. In each nomogram, every variable was assigned a special score according to the point scale (**Table 2**). The nomogram showed that the tumor subtype contributed the most to prognosis, followed by age and brain metastasis. By calculating the sum scores of each patient's clinical covariates, we can estimate the 1-year, 2-year, and 3-year OS and BCSS on the "total points" axis (**Figures 2A,B**).

The internal verification of the training set and external verification of the validation set were used to assess the credibility of the nomograms. The C-index of the OS nomogram was 0.701 in the training cohort and 0.699 in the validation cohort, and the C-index of the BCSS nomogram

TABLE 1 Demographics and clinicopathologic characteristics of the cohort with BCLM.

| Variables | Overall (*N* = 2537) | Training cohort (*N* = 1777) | Validation cohort (*N* = 760) | *P*-value |
|---|---|---|---|---|
| **Sex** | | | | 0.862 |
| Female | 2,497 (98.4%) | 1,748 (98.4%) | 749 (98.6%) | |
| Male | 40 (1.6%) | 29 (1.6%) | 11 (1.4%) | |
| **Age** | | | | 0.708 |
| <40 | 166 (6.5%) | 113 (6.4%) | 53 (7.0%) | |
| 40–59 | 956 (37.7%) | 682 (38.4%) | 274 (36.1%) | |
| 60–79 | 1,130 (44.5%) | 784 (44.1%) | 346 (45.5%) | |
| 80+ | 285 (11.2%) | 198 (11.1%) | 87 (11.4%) | |
| **Marital status** | | | | 0.573 |
| Married | 1,116 (44.0%) | 788 (44.3%) | 328 (43.2%) | |
| Unmarried | 1,421 (56.0%) | 989 (55.7%) | 432 (56.8%) | |
| **Race** | | | | |
| White | 1,833 (72.3%) | 1,294 (72.8%) | 539 (70.9%) | 0.6 |
| Black | 484 (19.1%) | 334 (18.8%) | 150 (19.7%) | |
| Other | 220 (8.7%) | 149 (8.4%) | 71 (9.3%) | |
| **Site** | | | | 0.676 |
| Inner | 277 (10.9%) | 200 (11.3%) | 77 (10.1%) | |
| Outer | 733 (28.9%) | 515 (29.0%) | 218 (28.7%) | |
| Other | 1,527 (60.2%) | 1,062 (59.8%) | 465 (61.2%) | |
| **Laterality** | | | | 0.516 |
| Left | 1,272 (50.1%) | 883 (49.7%) | 389 (51.2%) | |
| Right | 1,265 (49.9%) | 894 (50.3%) | 371 (48.8%) | |
| **Grade** | | | | 0.511 |
| I-II | 1,076 (42.4%) | 746 (42.0%) | 330 (43.4%) | |
| III-IV | 1,461 (57.6%) | 1,031 (58.0%) | 430 (56.6%) | |
| **AJCC_T** | | | | 0.655 |
| T1-2 | 957 (37.7%) | 665 (37.4%) | 292 (38.4%) | |
| T3-4 | 1,580 (62.3%) | 1,112 (62.6%) | 468 (61.6%) | |
| **AJCC_N** | | | | 0.789 |
| N0 | 523 (20.6%) | 369 (20.8%) | 154 (20.3%) | |
| N1-3 | 2,014 (79.4%) | 1,408 (79.2%) | 606 (79.7%) | |
| **Subtype** | | | | 0.248 |
| HR+/HER2− | 1,293 (51.0%) | 897 (50.5%) | 396 (52.1%) | |
| HR+/HER2+ | 471 (18.6%) | 346 (19.5%) | 125 (16.4%) | |
| HR−/HER2+ | 276 (10.9%) | 185 (10.4%) | 91 (12.0%) | |
| HR−/HER2− | 497 (19.6%) | 349 (19.6%) | 148 (19.5%) | |
| **Bone** | | | | 0.165 |
| No | 1,215 (47.9%) | 835 (47.0%) | 380 (50.0%) | |
| Yes | 1,322 (52.1%) | 942 (53.0%) | 380 (50.0%) | |
| **Brain** | | | | 0.199 |
| No | 2,307 (90.9%) | 1,607 (90.4%) | 700 (92.1%) | |
| Yes | 230 (9.1%) | 170 (9.6%) | 60 (7.9%) | |
| **Liver** | | | | 0.244 |
| No | 1,842 (72.6%) | 1,278 (71.9%) | 564 (74.2%) | |
| Yes | 695 (27.4%) | 499 (28.1%) | 196 (25.8%) | |
| **Surgery** | | | | 0.378 |
| No | 1,723 (67.9%) | 1,197 (67.4%) | 526 (69.2%) | |
| Yes | 814 (32.1%) | 580 (32.6%) | 234 (30.8%) | |
| **Chemotherapy** | | | | 0.174 |
| No/Unknown | 897 (35.4%) | 613 (34.5%) | 284 (37.4%) | |
| Yes | 1,640 (64.6%) | 1,164 (65.5%) | 476 (62.6%) | |
| **Radiation** | | | | 0.371 |
| No/Unknown | 1,762 (69.5%) | 1,244 (70.0%) | 518 (68.2%) | |
| Yes | 775 (30.5%) | 533 (30.0%) | 242 (31.8%) | |

For marital status, unmarried consists of single, divorced, separated, and widowed; For race, 'other' includes American Indian, AK Native, Asian, and Pacific Islander; For grade, Grade I means well-differentiated, grade II means moderately differentiated, grade III means poorly differentiated, Grade IV means undifferentiated or anaplastic.

was 0.708 in the training group and 0.697 in the validation group (Supplementary Table S2). In the training set, the area under the time-dependent ROC curve (AUC) of the nomogram to predict 1-, 2- and 3-year OS and BCSS ranged from 0.745 to 0.753 (Figures 3A,B). In the validation cohort, the AUC values of the nomogram to predict 1-, 2- and 3-year OS and BCSS ranged from 0.749 to 0.763 (Figures 3C,D). The calibration curves in both the training cohort and validation cohort showed good consistency between the model-based predictions and the actual observations (Figure 4).

## Risk stratification system

Based on the nomogram, we further established a risk classification and evaluated the impact of clinicopathological baseline data risk on the prognosis of patients. We calculated the sum scores of ten independent predictors (including age, marital status, race, laterality, grade, AJCC T stage, subtype, bone metastasis, brain metastasis, and liver metastasis), only demographic characteristics and disease characteristics were included. The median of the sum scores was set as the threshold. Above the median of predicted total scores was defined as high risk, as well below the median of predicted total scores was defined as low risk. For OS, BCLM patients were split into the low-risk group (scores < 147) and the high-risk group (scores ≥147). For BCSS, BCLM patients were separated into the low-risk group (scores < 139) and the high-risk group (scores ≥139). In the total cohort, the patients at low risk had better OS and BCSS compared with all BCLM patients, the BCLM patients at high risk showed worse OS and BCSS compared with all BCLM patients (Figures 5A,B). The Kaplan-Meier curves visibly differentiated the prognostic

TABLE 2 Multivariate Cox regression analysis for overall survival (OS) and breast cancer-specific survival (BCSS) of BCLM patients in the training cohort.

| Variables | OS | | | BCSS | | |
|---|---|---|---|---|---|---|
| | HR (95% CI) | *P*-value | Points | HR (95% CI) | *P*-value | Points |
| **Sex** | | | | | | |
| Female | – | – | – | – | – | – |
| Male | – | – | – | – | – | – |
| **Age** | | | | | | |
| <40 | Reference | | 0 | Reference | | 0 |
| 40–59 | 1.288 (1.005–1.651) | 0.0453 | 19 | 1.322 (1.019–1.715) | 0.0358 | 20 |
| 60–79 | 1.480 (1.155–1.897) | 0.0020 | 30 | 1.427 (1.099–1.854) | 0.0077 | 26 |
| 80+ | 2.458 (1.844–3.277) | 0.0000 | 68 | 2.264 (1.666–3.078) | 0.0000 | 60 |
| **Marital status** | | | | | | |
| Married | Reference | | 0 | Reference | | 0 |
| Unmarried | 1.319 (1.177–1.478) | 0.0000 | 21 | 1.2991.151–1.466) | 0.0000 | 19 |
| **Race** | | | | | | |
| White | Reference | | 14 | Reference | | 15 |
| Black | 1.228 (1.065–1.415) | 0.0047 | 30 | 1.184 (1.017–1.377) | 0.0293 | 27 |
| Other | 0.833 (0.678–1.023) | 0.0819 | 0 | 0.814 (0.653–1.014) | 0.0660 | 0 |
| **Site** | | | | | | |
| Inner | – | – | – | – | – | – |
| Outer | – | – | – | – | – | – |
| Other | – | – | – | – | – | – |
| **Laterality** | | | | | | |
| Left | Reference | | 0 | Reference | | 0 |
| Right | 1.160 (1.041–1.293) | 0.0074 | 11 | 1.163 (1.036–1.306) | 0.0104 | 11 |
| **Grade** | | | | | | |
| I-II | Reference | | 0 | Reference | | 0 |
| III-IV | 1.402 (1.236–1.590) | 0.0000 | 26 | 1.493 (1.304–1.709) | 0.0000 | 20 |
| **AJCC_T** | | | | | | |
| T1-2 | Reference | | 0 | Reference | | 0 |
| T3-4 | 1.307 (1.164–1.468) | 0.0000 | 20 | 1.354 (1.196–1.534) | 0.0000 | 22 |
| **AJCC_N** | | | | | | |
| N0 | – | – | – | – | – | – |
| N1-3 | – | – | – | – | – | – |
| **Subtype** | | | | | | |
| HR+/HER2− | Reference | | 18 | Reference | | 18 |
| HR+/HER2+ | 0.789 (0.669–0.930) | 0.0048 | 0 | 0.777 (0.652–0.926) | 0.0047 | 0 |
| HR−/HER2+ | 1.207 (0.982–1.485) | 0.0738 | 32 | 1.102 (0.881–1.377) | 0.3950 | 26 |
| HR−/HER2− | 2.937 (2.492–3.462) | 0.0000 | 100 | 3.051 (2.566–3.629) | 0.0000 | 100 |
| **Bone** | | | | | | |
| No | Reference | | 0 | Reference | | 0 |
| Yes | 1.355 (1.198–1.532) | 0.0000 | 23 | 1.3206 (1.1583–1.5057) | 0.0000 | 20 |
| **Brain** | | | | | | |
| No | Reference | | 0 | Reference | | 0 |
| Yes | 1.926 (1.618–2.294) | 0.0000 | 50 | 1.9160 (1.593–2.305) | 0.0000 | 48 |
| **Liver** | | | | | | |
| No | Reference | | 0 | Reference | | 0 |
| Yes | 1.644 (1.451–1.863) | 0.0000 | 38 | 1.778 (1.559–2.028) | 0.0000 | 42 |

(*continued*)

TABLE 2 Continued

| Variables | OS | | | BCSS | | |
|---|---|---|---|---|---|---|
| | HR (95% CI) | *P*-value | Points | HR (95% CI) | *P*-value | Points |
| **Surgery** | | | | | | |
| No | Reference | | 20 | Reference | | 21 |
| Yes | 0.770 (0.680–0.872) | 0.0000 | 0 | 0.747 (0.654–0.853) | 0.0000 | 0 |
| **Chemotherapy** | | | | | | |
| No/Unknown | Reference | | 35 | Reference | | 32 |
| Yes | 0.633 (0.556–0.721) | 0.0000 | 0 | 0.648 (0.563–0.745) | 0.0000 | 0 |
| **Radiation** | | | | | | |
| No/Unknown | – | – | – | – | – | – |
| Yes | – | – | – | – | – | – |

differences between the low-risk and the high-risk groups, indicating the excellent clinical utility of the nomograms.

## Kaplan Meier analyses of different treatments in stratified risk groups

According to stratified risk groups, we further investigated the survival benefit of primary site surgery, chemotherapy, and radiotherapy for BCLM patients. As illustrated in Figures 6A,D, primary site surgery remarkably prolonged OS of BCLM patients in both the low-risk ($P < 0.0001$) and high-risk groups ($P < 0.0001$). Furthermore, chemotherapy had a favorable effect on the OS of BCLM patients in both the low-risk ($P < 0.0001$) and high-risk groups ($P < 0.0001$) (Figures 6B,E). However, radiotherapy neither improved the OS of BCLM patients in the low-risk group ($P = 0.98$) nor improved the OS of those in the high-risk group ($P = 0.55$) (Figures 6C,F). The same outcomes could be observed for BCSS of BCLM patients as shown in Figure 7. The outcomes above showed that primary site surgery and chemotherapy are beneficial to BCLM patients, whether they are at low-risk or high-risk.

# Discussion

It is universally known that diverse clinicopathological parameters and molecular characteristics are closely related to clinical outcomes in BCLM patients. For example, a retrospective study reported that the prognosis of BCLM patients is dissatisfactory, with an 11-month median survival time in TNBC, and better outcomes of 31 months in HR +/HER2 + (4). BCLM patients who suffer the additional metastatic disease at distant sites (brain, liver, bone) obtain worse survival results compared to patients without distant metastases (8). For the complexity of the multivariate prognostic factors affecting the survival of BCLM patients, it was difficult to estimate the survival outcomes for BCLM patients. Therefore, we developed two prognostic nomograms to predict OS and BCSS for BCLM patients.

In the present study, age, marital status, race, laterality, grade, AJCC T stage, subtype, bone metastasis, brain metastasis, liver metastasis, surgery, and chemotherapy were found to be independent predictors of OS and BCSS. Chen S et al. have reported that age, race, marital status, pathological grade, molecular subtype, and extrapulmonary metastatic sites were survival risk factors for BCLM (5). In BCLM, age, black race, HR−/HER2 + subtype, triple-negative subtype, and higher grade had an adverse influence on the long-term prognosis of patients, while HR+/HER2 + subtype and marital status showed a favorable effect on the long-term survival of patients (4). These results were generally consistent with our reports. Additionally, we found that laterality is also a survival predictor for BCLM patients, and breast cancer on the left side has a better prognosis than breast cancer on the right side, which is not shown in other studies. The reason for this may be selection bias, and more studies and further prospective randomized trials with rigorous inclusion criteria are eagerly awaited to verify our results. In addition, the T stage was also associated with the prognosis of BCLM, and a lower T stage implied better survival. Furthermore, other factors mentioned above, primary site surgery and chemotherapy were also identified as significant predictors of prognosis.

Two nomograms were established to visualize the predictive survival of BCLM patients based on the results of multivariate Cox analysis. The nomograms in the present study could accurately estimate the prognosis of BCLM patients, which is helpful to the clinical management of patients. For the purpose of better understanding the use of the nomograms, we took a patient with BCLM as an example. A 50-year-old woman, married, white, right side of breast cancer, grade IV, AJCC T4, HR-/HER-, with lung metastases from breast

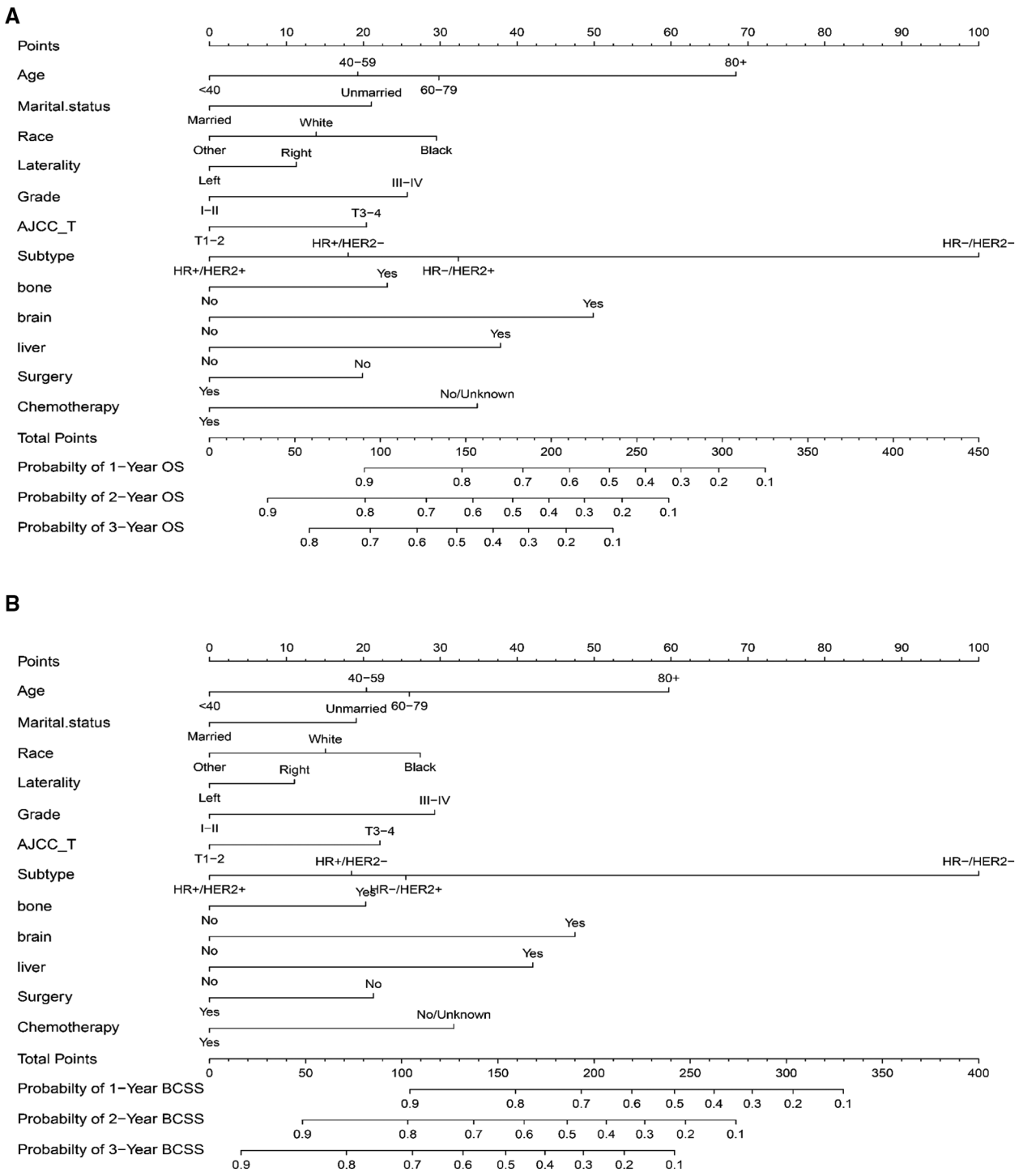


FIGURE 2
Nomograms for predicting 1-, 2-, and 3-year (A) overall survival (OS) and (B) breast cancer-specific survival (BCSS) for BCLM patients. HR, hormone receptor; HER2, Human epidermal growth factor receptor 2; AJCC, American Joint Committee on Cancer.

cancer, and no metastases beyond the lung, received surgery and chemotherapy, the patient had approximately 67%, 45%, and 28% of 1-, 2-, and 3-year overall survival probabilities, respectively. Traditionally, the main treatment for metastatic breast cancer is normally palliative care and supportive care, which aims at maintaining the quality of life and relieving

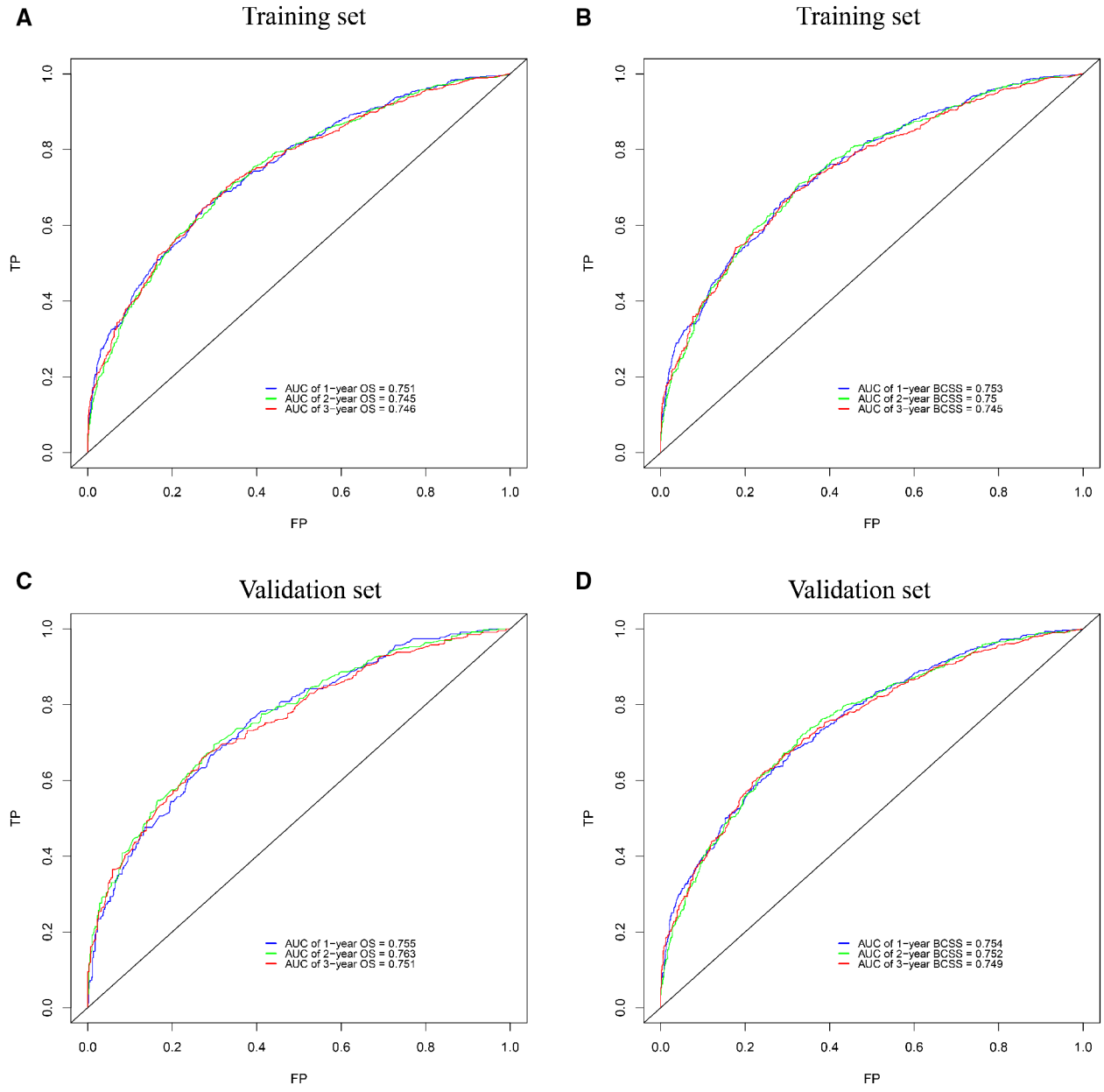


FIGURE 3
ROC curves for survival prediction of BCLM patients. **(A,B)** ROC curves of 1-, 2-, and 3-year overall survival (OS) and breast cancer-specific survival (BCSS) in the training set; **(C,D)** ROC curves of 1-, 2-, and 3-year OS and BCSS in the validation set. ROC: receiver operating characteristic, AUC: the area under the time-dependent ROC curve.

symptoms. An accurate survival estimation can assist clinicians and patients in making the most appropriate treatment plan, and was conducive to the rational utilization and allocation of medical resources. If the predicted survival rate is good, we can choose a more aggressive treatment strategy. If the predicted survival rate is poor, negative treatment methods such as palliative care and supportive care are more suitable for the patients, so as to avoid the side effects caused by aggressive treatment and improve the quality of life. Predicting the survival risk of BCLM patients can facilitate individualized treatment regimens, which is of great significance for improving the prognosis and quality of life for BCLM patients.

We used multiple methods to verify the clinical efficacy of the constructed nomograms. The predictive performance of the nomograms was evaluated by discrimination and

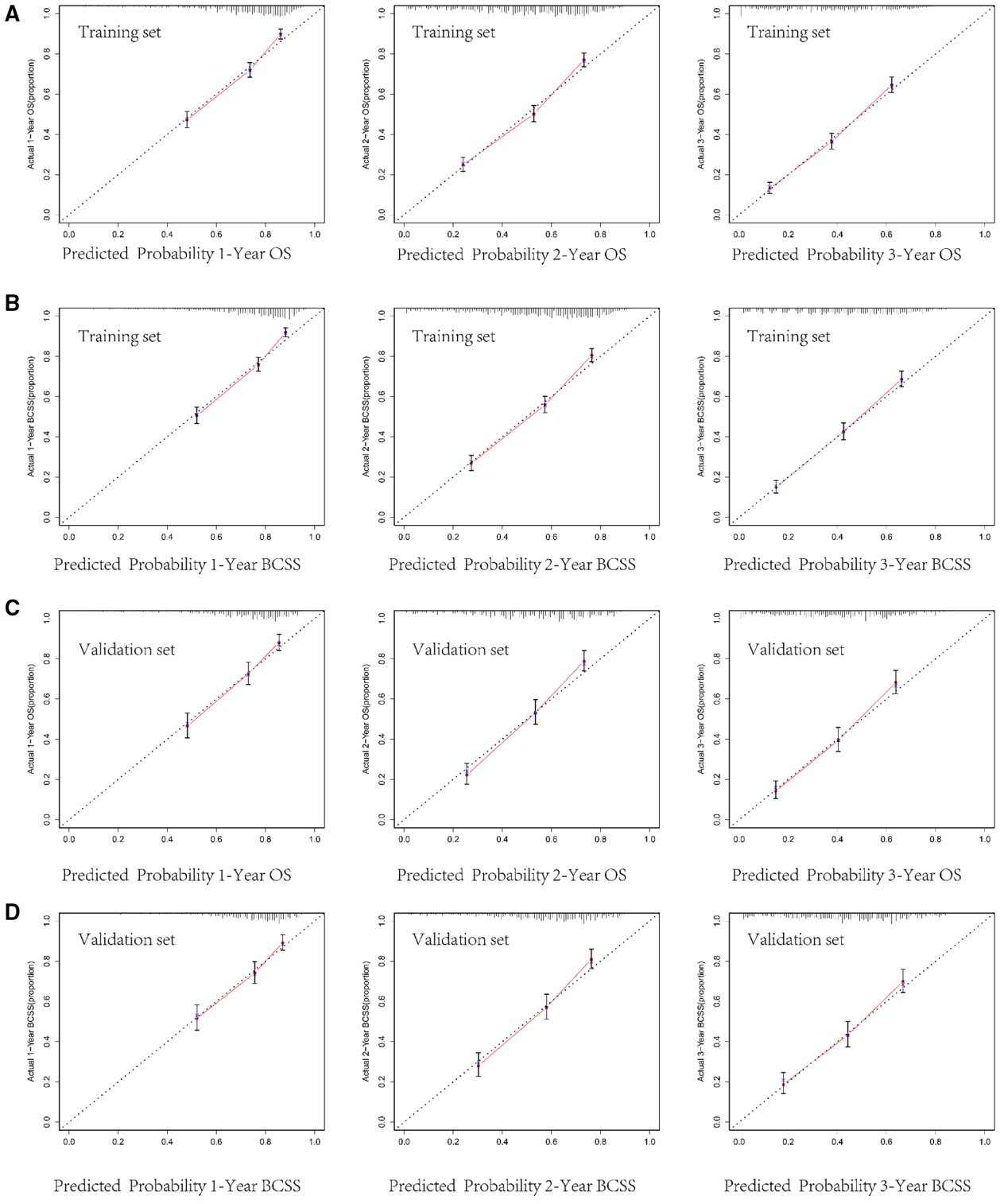


FIGURE 4
Calibration curves for predicting 1-, 2-, and 3-year overall survival (OS) and breast cancer-specific survival (BCSS) in the training cohort (A,B) and in the validation cohort (C,D).

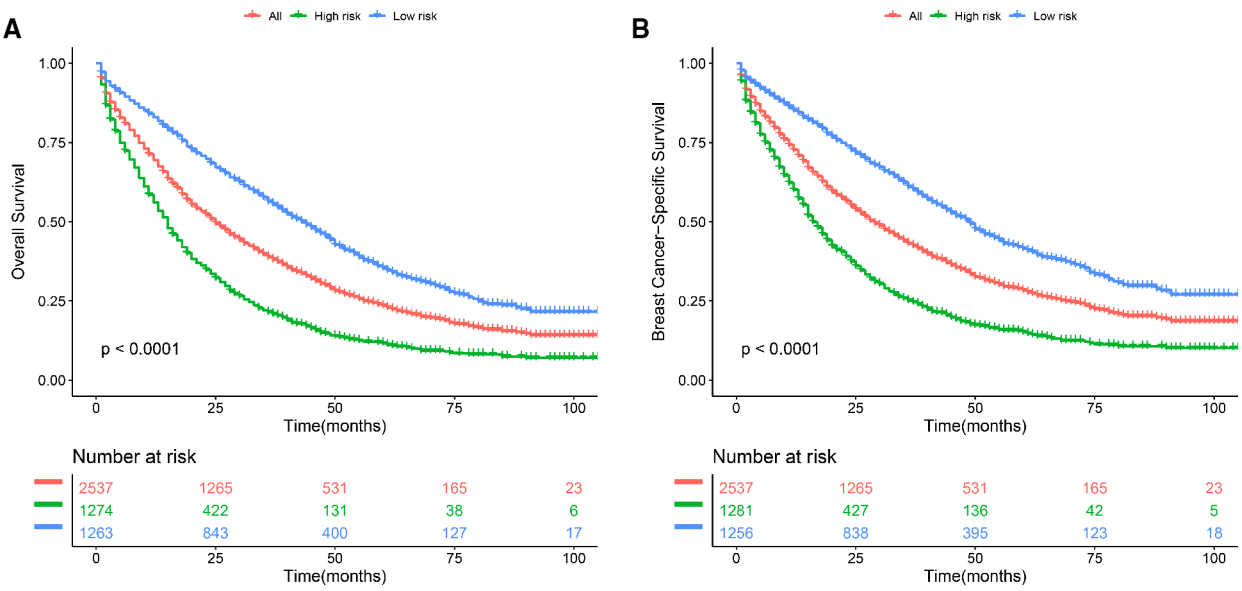


FIGURE 5
Kaplan–Meier curve of (**A**) overall survival (OS) and (**B**) breast cancer-specific survival (BCSS) to test the stratification system in the total cohort. All: all BCLM patients.

calibration internally and externally. The C-index was approximately 0.7, suggesting a good discrimination ability of the nomograms. The AUC values of 0.7 to 0.8 indicated that our nomograms showed great predictive ability for the prognosis of BCLM patients. The calibration curves showed excellent consistency between the actual observations and the predicted outcomes in predicting OS and BCSS, which guaranteed the reliability of the established nomograms. We also stratified the prognostic risk of BCLM patients based on nomograms. The significant difference in Kaplan–Meier curves among the low-risk and the high-risk groups confirmed the excellent predictive ability of the nomograms.

In our research, primary site surgery and chemotherapy could remarkably prolong OS and BCSS of BCLM patients no matter whether the patients were at low-risk or high-risk. Currently, chemotherapy, targeted therapy, and endocrine therapy are beneficial to the long-term survival of metastatic breast cancer and are the first-line treatment strategies for advanced breast cancer. But the role of primary site surgery (breast resection) in metastatic breast cancer is still controversial. For stage IV breast cancer, resection of the primary tumor can reduce tumor burden and control cancer-related symptoms. Conversely, it has also been reported that primary site surgery may accelerate the emergence of distant metastasis by inducing angiogenesis and proliferation of distant dormant micrometastases (9). In terms of existing evidence, some studies showed that breast cancer patients with bone metastasis alone can benefit from resection of the primary tumor, while patients with visceral metastasis do not (10–12). However, another study showed that surgery is related to better OS in breast cancer patients with single metastasis to the liver, lung, or brain (13). The NCCN guidelines for breast cancer suggest that surgery at the primary site is not recommended except for patients who can benefit from initial systemic therapy (14). Radiotherapy, as a local treatment, is often used as adjuvant therapy for breast cancer receiving breast-conserving surgery. Radiotherapy also has been a palliative treatment strategy that aims to control tumor progression and suppress tumor-related symptoms for cancer patients with metastatic diseases. Radiotherapy had improvement in locoregional recurrence, however, this does not translate into an advantage in the overall survival of early-stage breast cancer patients (15, 16). Few high-evidence studies like randomized controlled trials were conducted to investigate the effect of radiotherapy among *de novo* stage IV breast cancer patients so far. Our results showed that radiotherapy did not improve the survival outcomes of BCLM patients. But as an effective strategy in controlling local lesions, radiotherapy is often used in combination with drug therapy for advanced breast cancer. Our results could provide some basis for the treatment choice of patients with BCLM to some extent.

Inevitably, some limitations were in this research. First, there is no data on the different options of the systemic treatment used. Endocrine therapy and targeted therapy play vital roles in the treatment of metastatic or advanced breast

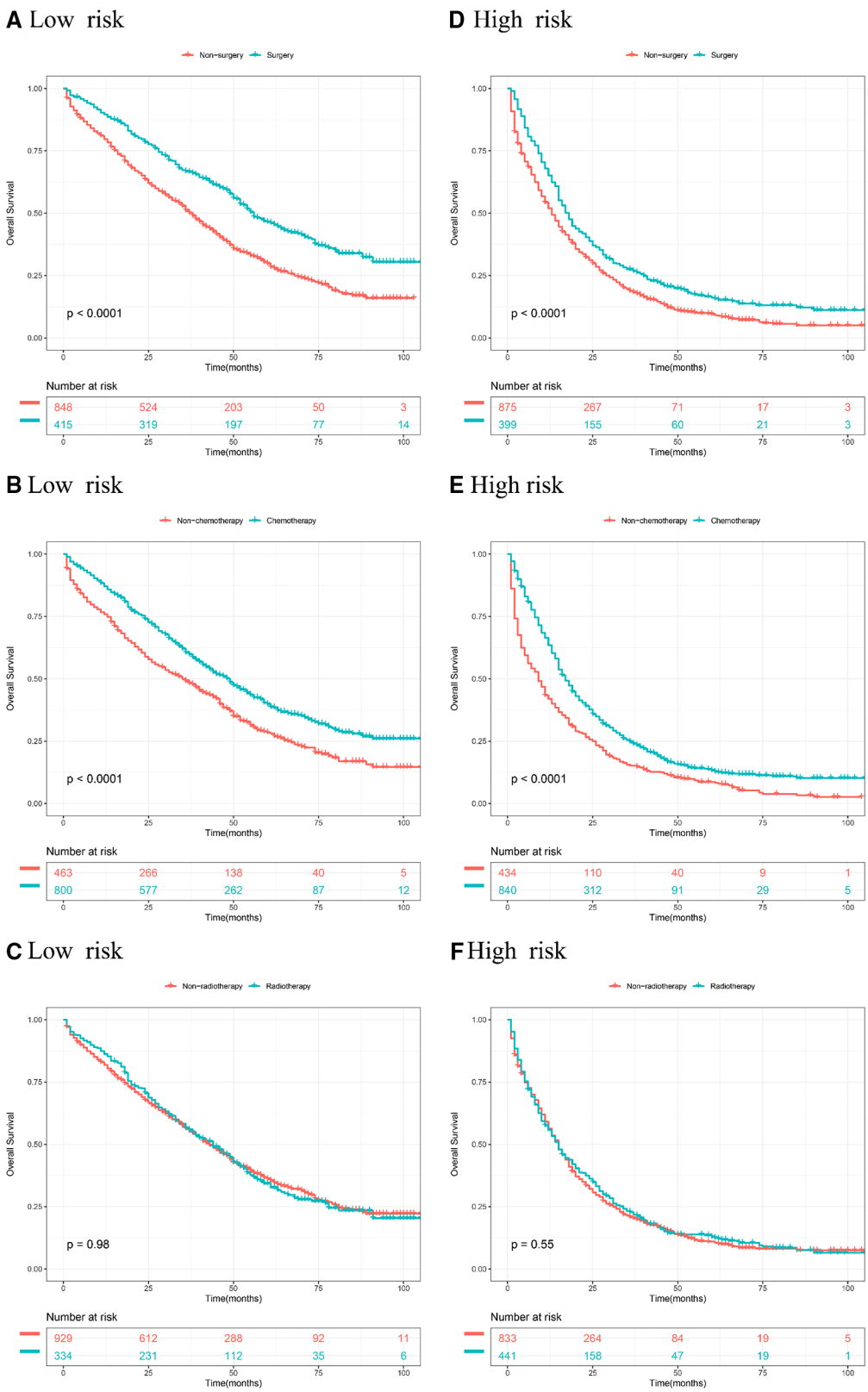


FIGURE 6
Kaplan–meier curves of different treatments for risk stratification in terms of OS. Kaplan–Meier curves of primary surgery in the low-risk group (**A**) and high-risk group (**B**); Kaplan–Meier curves of chemotherapy in the low-risk group (**C**) and the high-risk group (**D**); Kaplan–Meier curves of radiotherapy in the low-risk group (**E**) and the high-risk group (**F**).

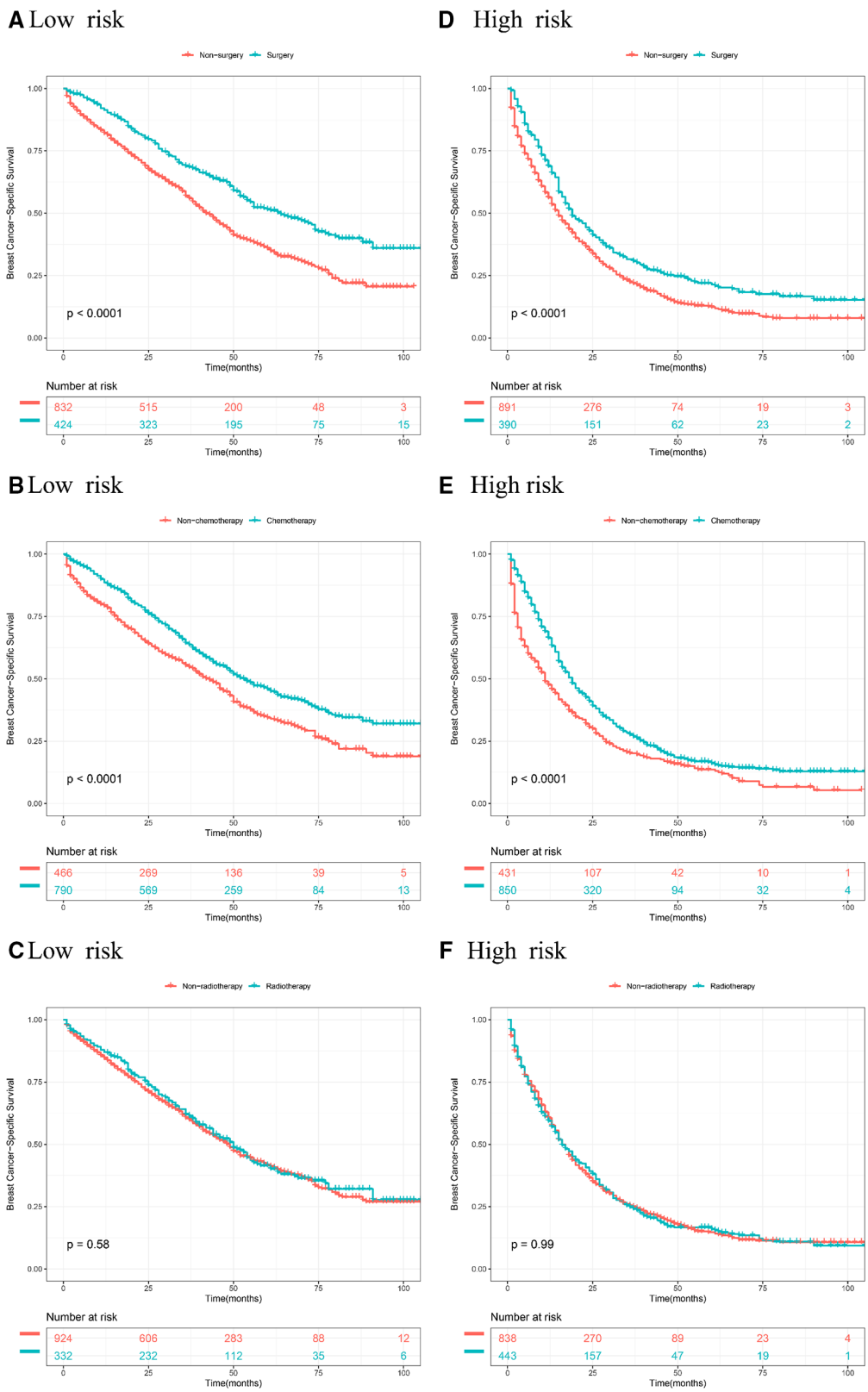


FIGURE 7
Kaplan–Meier curves of different treatments for risk stratification in terms of BCSS. Kaplan–Meier curves of primary surgery in the low-risk group (**A**) and high-risk group (**B**); Kaplan–Meier curves of chemotherapy in the low-risk group (**C**) and the high-risk group (**D**); Kaplan–Meier curves of radiotherapy in the low-risk group (**E**) and the high-risk group (**F**).

cancer, but the information was not recorded in the SEER database, leading to the deviation of patient survival prediction to some extent. Second, information about lung metastases was absent, such as the data on the type of metastatic lesions to the lungs (single, multiple). A large number of retrospective studies have presented obvious benefits for BCLM patients who undergo pulmonary metastasectomy (17–20). The number of lung metastasis influences the choice of the further procedure because a single lesion to the lung was possibly to select surgical excision, and the lack of relevant information may affect the accuracy of the model in predicting survival. Third, other metastatic sites that may affect the prognosis of metastatic breast cancer, such as the peritoneum, other internal organs, or skin, were not collected in this study. Fourth, we do not take the general condition of patients into account owing to the inherent biases in the SEER database (21), which often affects the therapeutic possibilities. Finally, although our models showed excellent predictive performance, they had not been validated in other centers or databases.

In conclusion, age, marital status, race, laterality, grade, AJCC T stage, subtype, bone metastasis, brain metastasis, liver metastasis, surgery, and chemotherapy were identified as independent prognostic indicators for BCLM. The first prognostic nomogram created for BCLM can excellently predict individual survival and assist clinicians in optimizing individualized treatment strategies for BCLM patients.

## Data availability statement

Publicly available datasets were analyzed in this study. This data can be found here: https://seer.cancer.gov/.

## Ethics statement

The data analyzed in the present study was collected from the SEER database, and the identifiable information of patients was undisclosed; therefore, this article was exempted from approval by the Institutional Review Board.

## Author contributions

WWY designed the study, obtained the data, conducted data analysis, and drafted the manuscript. LJJ participated in the data analysis and revision of the manuscript. CYQ supervised the project and managed the Figures and tables. XXF and HLQ supervised the project. WXL revised the manuscript. GJ supervised the project and revised the manuscript. All authors contributed to the article and approved the submitted version.

## Conflict of interest

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

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## Supplementary material

The Supplementary Material for this article can be found online at: https://www.frontiersin.org/articles/10.3389/fsurg.2022.939132/full#supplementary-material.

## References

1. Bray F, Ferlay J, Soerjomataram I, Siegel RL, Torre LA, Jemal A. Global cancer statistics 2018: gLOBOCAN estimates of incidence and mortality worldwide for 36 cancers in 185 countries. *CA Cancer J Clin.* (2018) 68:394–424. doi: 10.3322/caac.21492

2. DeSantis CE, Ma J, Gaudet MM, Newman LA, Miller KD, Goding Sauer A, et al. Breast cancer statistics, 2019. *CA Cancer J Clin.* (2019) 69:438–51. doi: 10.3322/caac.21583

3. Wang R, Zhu Y, Liu X, Liao X, He J, Niu L. The clinicopathological features and survival outcomes of patients with different metastatic sites in stage IV breast cancer. *Bmc Cancer.* (2019) 19:1091. doi: 10.1186/s12885-019-6311-z

4. Xiao W, Zheng S, Liu P, Zou Y, Xie X, Yu P, et al. Risk factors and survival outcomes in patients with breast cancer and lung metastasis: a population-based study. *Cancer Med (Malden, MA).* (2018) 7:922–30. doi: 10.1002/cam4.1370

5. Chen S, Yang J, Liu Y, You H, Dong Y, Lyu J. Prognostic factors and survival outcomes according to tumor subtype in patients with breast cancer lung metastases. *Peerj.* (2019) 7:e8298. doi: 10.7717/peerj.8298

6. Balachandran VP, Gonen M, Smith JJ, DeMatteo RP. Nomograms in oncology: more than meets the eye. *Lancet Oncol.* (2015) 16:e173–80. doi: 10.1016/S1470-2045(14)71116-7

7. Xiong Y, Shi X, Hu Q, Wu X, Long E, Bian Y. A nomogram for predicting survival in patients with breast cancer liver metastasis: a population-based study. *Front Oncol*. (2021) 11:600768. doi: 10.3389/fonc.2021.600768

8. Guo Y, Arciero CA, Jiang R, Behera M, Peng L, Li X. Different breast cancer subtypes show different metastatic patterns: a study from A large public data base. *Asian Pac J Cancer Prev*. (2020) 21:3587–93. doi: 10.31557/APJCP.2020.21.12.3587

9. Baum M, Demicheli R, Hrushesky W, Retsky M. Does surgery unfavourably perturb the "natural history" of early breast cancer by accelerating the appearance of distant metastases? *Eur J Cancer*. (2005) 41:508–15. doi: 10.1016/j.ejca.2004.09.031

10. Rhu J, Lee SK, Kil WH, Lee JE, Nam SJ. Surgery of primary tumour has survival benefit in metastatic breast cancer with single-organ metastasis, especially bone. *Anz J Surg*. (2015) 85:240–4. doi: 10.1111/ans.12548

11. Huang Z, Tan Q, Qin Q, Mo Q, Wei C. Impact of primary site surgery on survival of patients with de novo stage IV breast cancer. *Cancer Manag Res*. (2021) 13:319–27. doi: 10.2147/CMAR.S280470

12. Yu Y, Hong H, Wang Y, Fu T, Chen Y, Zhao J, et al. Clinical evidence for locoregional surgery of the primary tumor in patients with De Novo stage IV breast cancer. *Ann Surg Oncol*. (2021) 28(9):5059–70. doi: 10.1245/s10434-021-09650-3

13. Bilani N, Elson L, Liang H, Elimimian EB, Nahleh Z. Effect of surgery at primary and metastatic sites in patients with stage IV breast cancer. *Clin Breast Cancer*. (2021) 21:170–80. doi: 10.1016/j.clbc.2020.08.008

14. Gradishar WJ, Moran MS, Abraham J, Aft R, Agnese D, Allison KH, et al. NCCN Guidelines insights: breast cancer, version 4.2021. *J Natl Compr Canc Netw*. (2021) 19:484–93. doi: 10.6004/jnccn.2021.0023

15. Hughes KS, Schnaper LA, Bellon JR, Cirrincione CT, Berry DA, McCormick B, et al. Lumpectomy plus tamoxifen with or without irradiation in women age 70 years or older with early breast cancer: long-term follow-up of CALGB 9343. *J Clin Oncol*. (2013) 31:2382–7. doi: 10.1200/JCO.2012.45.2615

16. Kunkler IH, Williams LJ, Jack WJ, Cameron DA, Dixon JM. PRIME II. Breast-conserving surgery with or without irradiation in women aged 65 years or older with early breast cancer (PRIME II): a randomised controlled trial. *Lancet Oncol*. (2015) 16:266–73. doi: 10.1016/S1470-2045(14)71221-5

17. Endoh M, Shiono S, Yamauchi Y, Mun M, Ikeda N, Hashimoto H, et al. Pulmonary metastasectomy for pulmonary metastasis of breast cancer has a limited prognostic impact: a multi-institutional retrospective analysis. *J Thorac Dis*. (2020) 12:6552–62. doi: 10.21037/jtd-20-1788

18. Kycler W, Laski P. Surgical approach to pulmonary metastases from breast cancer. *Breast J*. (2012) 18:52–7. doi: 10.1111/j.1524-4741.2011.01176.x

19. Chen F, Fujinaga T, Sato K, Sonobe M, Shoji T, Sakai H, et al. Clinical features of surgical resection for pulmonary metastasis from breast cancer. *Eur J Surg Oncol*. (2009) 35:393–7. doi: 10.1016/j.ejso.2008.05.005

20. Bilani N, Yaghi M, Singh Jabbal I, Elson L, Elimimian EB, Liang H, et al. Survival benefit of a combined surgical approach in patients with metastatic breast cancer. *J Surg Oncol*. (2021) 124(8):1235–41. doi: 10.1002/jso.26656

21. Doll KM, Rademaker A, Sosa JA. Practical guide to surgical data sets: surveillance, epidemiology, and End results (SEER) database. *Jama Surg*. (2018) 153:588–9. doi: 10.1001/jamasurg.2018.0501

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# An alternative palliative surgical method for advanced malignant obstructive jaundice: Laparoscopic bridge choledochoduodenostomy

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**Background:** This study introduces an alternative palliative surgical procedure called laparoscopic bridge choledochoduodenostomy (LBCDD) for patients with advanced malignant obstructive jaundice (AMOJ).
**Methods:** Patients with AMOJ who had LBCDD between January 2017 and August 2021 were identified from databases of two institutions in China.
**Results:** A total of 35 patients (male 12; female 23) with an average age of 64 years were enrolled. The average diameter of the tumor is 4.24 cm. All patients undertook LBCDD within an average operation time of 75 min with a mean blood loss of 32 ml. One patient had controlled bile leakage after the operation and two developed surgical site infection involving the epigastric orifices. All of them were solved by conservative treatment. All patients were discharged smoothly after an average hospital stay of 5.5 days, and no conversion to open surgery was required.
**Conclusions:** LBCDD is a safe and efficient palliative surgery, which has a good therapeutic effect on patients with AMOJ.

KEYWORDS
obstructive jaundice, choledochoduodenostomy, laparoscopic, biliary drainage, bilioenteric anastomosis

## Introduction

Malignant obstructive jaundice can cause many adverse events including severe cholangitis, lower the quality of life, and increase mortality, which can occur following pancreatic cancer, hilar cholangiocarcinoma, and periampullary carcinoma (1–4). For advanced malignant obstructive jaundice (AMOJ) with no chance for radical cure, although combined treatment and local treatment are indispensable (5–7), effective and reliable biliary drainage is the most important palliative treatment (1–4, 8, 9).

Abbreviations
AMOJ, advanced malignant obstructive jaundice; LBCDD, laparoscopic bridge choledochoduodenostomy; OS, overall survival; PTBD, percutaneous transhepatic biliary drainage; EBD, endoscopic biliary drainage; ASA, American Anesthesiology Association; CBD, common bile.

Percutaneous transhepatic biliary drainage (PTBD), endoscopic biliary drainage (EBD) and bilioenteric anastomosis are the commonly used clinical methods for AMOJ at present (2, 9, 10). As an external biliary drainage, PTBD may lead to nutritional loss, gastrointestinal dysfunction, and a series of stable immune systems due to the long-term loss of large amounts of bile (3, 9, 11–13). Moreover, with low compliance, tube outside the body may cause psychological burden (3, 9, 11). As internal biliary drainage, EBD and bilioenteric anastomosis can avoid external biliary drainage problems. However, EBD cannot be applied to cases of severe biliary obstruction (4, 9, 11). Bilioenteric anastomosis is considered to be the most effective palliative treatment for advanced malignant obstructive jaundice. However, for malignant cases involving high bile duct position, some patients cannot complete bilioenteric anastomosis because of the short normal bile duct, such as advanced hilar cholangiocarcinoma (8, 14, 15). Therefore, we present a new laparoscopic surgical procedure, which bridges the common bile duct and duodenum through a T-tube and constructs a bile internal drainage. The new surgical procedure was called laparoscopic bridge choledochoduodenostomy (LBCDD), as the T-tube acted as a bridge for bile drainage in this surgical procedure. This surgical method may provide an alternative way of internal bile drainage for AMOJ. The present study is to assess the efficacy, safety, and feasibility of this novel surgical procedure.

# Methods

## General information and grouping

Patients with AMOJ who had LBCDD between January 2017 and August 2021 were identified from the electronic database of Central Hospital of Dengzhou and Henan Provincial People's Hospital. Inclusion criteria included patients with obstructive jaundice due to bile duct and pancreatic, ampullary, or duodenal malignancy who had lost the opportunity for radical or transformational therapy. The current treatment for these patients is mainly to relieve jaundice, and the patients or their family members refused external drainage and strongly required internal drainage. The present study was approved by the ethics committee of the hospitals. All patients signed the informed consent. A total of 35 patients with AMOJ who had LBCDD were enrolled. Fifteen cases of pancreatic carcinoma, 12 cases of terminal bile duct carcinoma, 5 cases of ampullary carcinoma, and 3 cases of duodenal adenocarcinoma were involved in this study. All diagnosis was confirmed by B-ultrasound, computed tomography (CT), magnetic resonance imaging (MRI), or magnetic resonance cholangiopancreatography (MRCP). Among them, 21 patients also had preoperative endoscopic procedures. Because most of the patients did not receive tumor resection or puncture biopsy, their diagnosis was clinical diagnosis without histopathological confirmation. Each operation was evaluated repeatedly on the basis of preoperative data and intraoperative situations under laparoscopy to ensure surgical safety. All patients had a good clinical record and were identified as being in an advanced stage, losing the chance of radical surgery. Recorded data such as symptoms, comorbidities, blood imaging studies, investigations, surgical data, postoperative variables, and follow-up data were collected. Continuous variables are represented by median values. The Charlson Comorbidity Index (CCI) was used to define the severity of comorbid conditions. Since patients' readmission to hospital may be strongly influenced by factors other than their condition, we did not count readmissions for evaluation.

## Positions of trocars and trimming of T-tube of LBCDD

Patients were placed in supine position or slightly inclined to the left on the operation table. After performing general anesthesia and intubation, the operating area was then disinfected. A 10 mm incision was first made at the right edge of the umbilicus and a 10 mm trocar was placed. Then, the laparoscope was placed after the pneumoperitoneum was constructed. Under the guidance of the laparoscope, two trocars for surgical instruments were placed below the xiphoid process (10 or 12 mm) and 2 cm below the costal margin of the right upper quadrant along the median line of the clavicle (5 mm), respectively (**Figure 1A**). If assistance is required, a trocar (5 mm) can also be placed along the midline of the clavicle 2 cm below the costal margin of the right upper quadrant. The T-tube serves as a bridge for bile from the common bile duct into the duodenum, retaining a length of 10–12 cm to ensure that the distal T-tube of the duodenum can pass through the duodenal papilla (**Figure 1B**).

## Surgical procedure of LBCDD and relevant precautions

After the exploration of the abdominal cavity, the common bile duct was exposed first (**Figure 2A**). An opening was made in the duodenum below the common bile duct, and a presutured double-layer suture was performed around (**Figure 2B**). The trimmed T-tube was placed in the common bile duct and fixed (**Figure 2C**). After the distal end of the T-tube was placed in duodenum through the open (**Figure 2D**), the presutured double-layer suture line was tightened (**Figure 2E**). Then, the adjacent greater omentum tissue was pulled to cover the T-tube (**Figure 2F**).

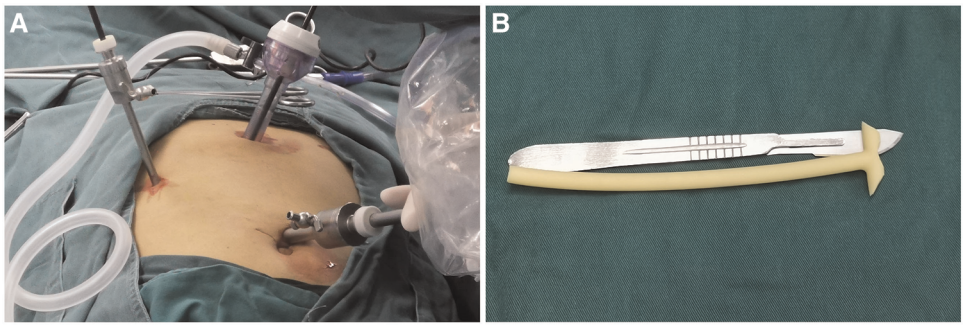


FIGURE 1
Positions of trocars and trimming of T-tube. Patients were placed in the supine position or slightly inclined to the left on the operation table. After general anesthesia and intubation, the operating area was then disinfected. A 10 mm incision was first made at the right edge of the umbilicus and a 10 mm trocar was placed. Then, the laparoscope was placed after the pneumoperitoneum was constructed. Under the guidance of the laparoscope, two trocars for surgical instruments were placed below the xiphoid process (10 or 12 mm) and 2 cm below the costal margin of the right upper quadrant along the median line of the clavicle (5 mm), respectively (**A**). If assistance is required, a trocar (5 mm) can also be placed along the midline of the clavicle 2 cm below the costal margin of the right upper quadrant. The T-tube serves as a bridge for bile from the common bile duct into the duodenum, retaining a length of 10–12 cm to ensure that the distal T-tube of the duodenum can pass through the duodenal papilla (**B**).

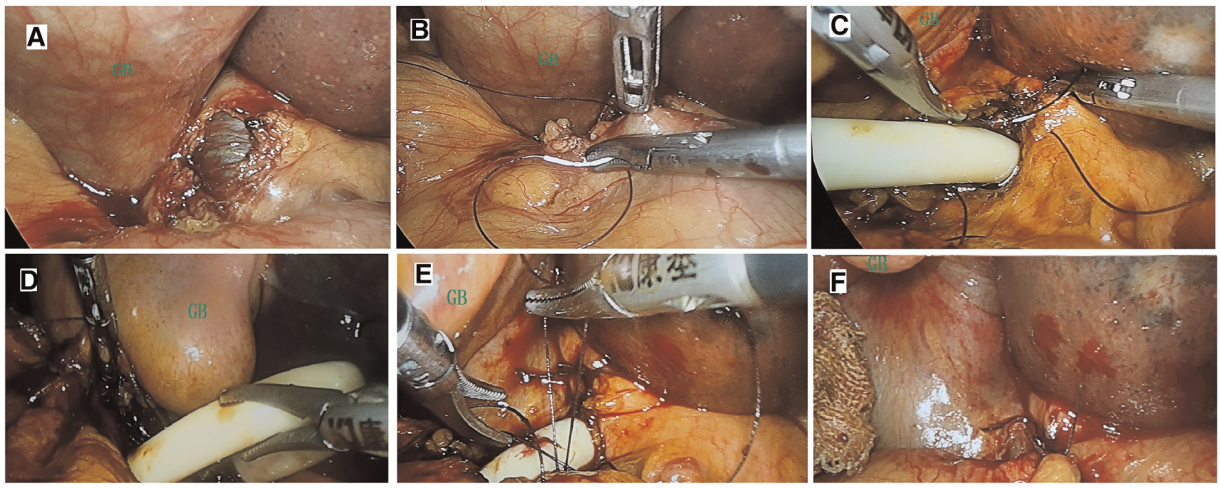


FIGURE 2
Surgical procedure of laparoscopic bridge choledochoduodenostomy. After the exploration of the abdominal cavity, the common bile duct was exposed first (**A**). An ostomy was made in the duodenum below the common bile duct, and a presutured double-layer suture was performed around (**B**). The trimmed T-tube was placed in the common bile duct and fixed (**C**). After the distal end of the T-tube was placed in duodenum through the ostomy (**D**), the presutured double-layer suture line was tightened (**E**). Then the adjacent greater omentum tissue was pulled to cover the T-tube (**F**). GB, gall bladder.

The surgical procedure can be simplified into three steps. The first step is to place a trimmed T-tube into the common bile duct and suture it for fixation. The second step is to place the distal end of the T-tube into the duodenal open and suture it for fixation. Then, adjacent tissues such as the greater omentum can be used to cover the exposed portion of the T-tube. It should be emphasized that the opening of the bile duct and duodenum should be as close together as possible and that the length of the T-tube in the duodenal lumen should extend beyond the duodenal papilla.

During the operation, to prevent bile or intestinal contents leaking into the abdominal cavity from the cutting open, we usually put an aspirator in the precut open position before incision, which can suck up the leaked bile or intestinal contents and minimize the abdominal pollution. Prophylactic application of the second-generation cephalosporin was

applied for 24 h postoperatively, which could be extended to 48 h for individual patients according to intraoperative conditions.

## Statistical analysis and follow-up

The cumulative summation (CUSUM) test was applied for the quantitative estimation of the learning curve (plotting the operation time and blood loss, and determination of the case number to achieve mastery) as described (16). Continuous variables were presented as mean ± SD and mean (range). Follow-up was performed by trained investigators through telephone calls, by recording the consultations of patients at the outpatient clinic every 2 weeks for 2 months postoperatively.

# Results

A total of 35 patients were enrolled, including 12 males and 23 females with a mean age of 64 (±10.65) years. The average body mass index of all patients was 26.15 (±3.5). The average diameter of the tumor is 4.24 (±1.11) cm, with a minimum of 3.5 cm and a maximum of 9.5 cm. Among them, 27 patients have a diameter of over 4 cm. All operations were performed within an average operating time of 75 (45–120) min with a mean blood loss of 32 (5–150) ml. The range of preoperative total bilirubin of all patients was between 135.1–632.5 mol/L, with a mean value of 241.24 ± 101.55 mol/L. Patients who developed comorbidities were kept in the ICU for 1 day after the operation. There was one patient who developed a controlled bile leak and two had surgical site infection (SSI) involving the epigastric port. All of them were resolved through a conservative way. The drain tube was removed 3 days postoperatively after a routine abdominal imaging examination, except the cases who had bile leak. There are no postoperative mortalities. All the patients were discharged smoothly with a mean hospital stay of 5.5 days, and no conversion to open surgery was required. During the mean follow-up duration of 14 (±4.3) months, no anastomose-related long-term complications have been found, which include strictures, cholangitis, or pancreatitis (**Table 1**). After operation, 29 patients received further chemotherapy and 8 accepted radiotherapy. By the end of December 2021, 29 patients had died, of which 1 patient died of gastrointestinal bleeding, and the others died of malignant fluid and systemic failure caused by the tumor. The median survival was 8.2 (±4.1) months. All patients were followed up and the results showed that total bilirubin had fallen below 50 mol/L in all patients 2 weeks after surgery.

Based on a visual analysis of the learning curve, a peak was noted in the 13th case (detailed information is listed in the **Supplementary Material**). Therefore, case 13 was defined as the learning-curve cutoff point regarding surgical time, blood loss, and complications after which the learning curve declined.

TABLE 1 Patient and surgery characteristics.

| Variable value | Value (mean ± SD) |
|---|---|
| Age (years) | 64 ± 10.65 |
| Sex | |
| Male | 12 (34.3%) |
| Female | 23 (65.7%) |
| Diagnosis (cases) | |
| Pancreatic carcinoma | 15 (42.86%) |
| Terminal bile duct carcinoma | 12 (34.29%) |
| Ampullary carcinoma | 5 (14.29%) |
| Duodenal adenocarcinoma | 3 (8.57%) |
| Body mass index (kg/m$^2$) | 26.15 ± 3.5 |
| CBD diameter (cm) | 1.5 ± 0.7 |
| Total bilirubin (mol/L) | 241.24 ± 101.55 |
| ALT (U/L) | 86 ± 107 |
| Tumor diameter (cm) | 4.24 ± 1.11 |
| AJCC stage | |
| III | 16 (45.9%) |
| IV | 19 (54.1%) |
| Charlson comorbidity index | |
| 0 | 27 (77.1%) |
| 1–3 | 8 (22.9%) |
| Operative time (min) | 75 (45–120) |
| Blood loss (ml) | 32.0 (5–150) |
| Complication | 3 (8.6%) |
| Bile leak | 1 (2.9%) |
| SSI-superficial | 2 (5.7%) |
| Hospital stays (days) | 5.5 ± 2.5 |

SD, standard deviation; CBD, common bile duct; SSI, surgical site infection.

# Discussion

Our study showed that the majority of AMOJ patients were elderly (64 ± 10.65 years), and females were 1.92 times as many as males. Since there was no opportunity of radical surgery for AMOJ patients, solving jaundice was the most important way to prolong life and improve their life quality. Most of the patients have a large tumor above 4 cm, which severely compacts or infiltrates the bile duct, making EBD impossible to perform. Bilioenteric anastomosis is reported including choledoduodenostomy and choledojejunostomy. This procedure could not be performed in patients enrolled in this study, mainly because the high bile duct was invaded by the tumor, and there was no sufficient length of normal bile duct for the anastomosis (17–19). Moreover, bilioenteric anastomosis has the risk of complications such as anastomotic leak and strictures (14, 15, 19, 20).

LBCDD is a novel internal drainage procedure, which avoids a series of external drainage-related complications such as weakened immunity and impaired gastrointestinal function caused by chronic and massive bile loss. In addition, with a high degree of compliance, LBCDD does not need to wear any tubes outside the body. LBCDD applied T-tube to drainage bile from common bile duct to the duodenum. A T was tube used as a bridge, which establish a channel between bile duct and duodenum. The T-tube length was controlled in 10–12 cm, so as to cross the duodenal papilla, which ensures that various digestive enzymes are activated away from the duodenal opening. Moreover, we used the greater omentum to cover the T-tube between the bile duct and duodenum. Those measures have effectively reduced the risk of anastomotic leaks. Duodenal leak is considered dreaded when we begin the procedure; however, none of the cases had this complication. Although the published leak rate of choledojejunostomy is 2%–7% (14, 15, 17), there is only one case of biliary leakage. In addition, there are two cases of surgical site infection. All of them occurred in the early stages of our learning curve. According to the follow-up data, the total bilirubin of all patients had fallen below 50 mol/L 2 weeks after surgery, and no delayed postoperative complications such as cholangitis, pancreatitis, and strictures occurred. Therefore, for patients with AMOJ who cannot be treated with EBD or bilioenteric anastomosis, as a safe surgical procedure, LBCDD may be an alternative for internal bile drainage.

In addition, the operation process of the present operation is simple and the operation time is short. Most of them can be completed around 1 h in the later stage of the term curve (after the learning-curve cutoff point of the 13th case), with an average operative time of 75 (±31) min. On one hand, the simplified surgical procedures can reduce the complications related to the operation. On the other hand, it also can reduce the operation cost and speed up postoperative recovery. The patient can have a liquid diet on the second day after the operation. Early eating can improve patient's in-patient experience and satisfaction, as well as ensure the patient's smooth postoperative recovery. The current study reported a comparable short hospital stay with a median length of 5.5 days.

Our study shows that LBCDD, as a novel surgical procedure, is a safe and efficient treatment for AMOJ. Compared with bilioenteric anastomosis, LBCDD does not need to cut the small intestine; it has a simpler surgical procedure, with less bleeding risk, requires no expensive supplies, and is more physiological. Therefore, LBCDD is worthy of recommendation. Since our study enrolled only 35 patients, the number is small, and the implementation of this technique requires sophisticated laparoscopic techniques; the replication of similar results may not be achieved during the early stages of performing this procedure. Moreover, this procedure requires an opening in the duodenum, there is a theoretical possibility of duodenal leakage for inexperienced physicians or patients with poor postoperative management. However, avoiding a series of external drainage-related shortcomings and with a high degree of compliance, LBCDD is a safe and simple operation, which can reduce the operation cost and speed up postoperative recovery. We would like to suggest LBCDD as an alternative option.

## Conclusion

LBCDD is a safe and efficient palliative surgery, which has a good therapeutic effect on patients with AMOJ.

## Data availability statement

The raw data supporting the conclusions of this article will be made available by the authors, without undue reservation.

## Ethics statement

Ethical approval was obtained from the respective institutional review boards of the Ethics Committee of Henan Provincial People's Hospital and Central Hospital of Dengzhou. All patients provided written informed consent to participate in this study.

## Author contributions

Each author took part in the design of the study, contributed to data collections, and participated in writing the manuscript, and all agree to accept equal responsibility for accuracy of the contents of this paper. The authors read and approved the final manuscript.

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## Conflict of interest

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

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## Supplementary material

The Supplementary Material for this article can be found online at: https://www.frontiersin.org/articles/10.3389/fsurg.2022.1056093/full#supplementary-material.

## References


1. Li J, Zhuo S, Chen B, Liu Y, Wu H. Clinical efficacy of laparoscopic modified loop cholecystojejunostomy for the treatment of malignant obstructive jaundice. *J Int Med Res*. (2020) 48(2):300060519866285. doi: 10.1177/0300060519866285

2. Pan T, Li MA, Mu LW, Zhu D, Qian JS, Li ZR. Stent placement with iodine-125 seeds strand effectively extends the duration of stent patency and survival in patients with unresectable malignant obstructive jaundice. *Scand J Gastroenterol*. (2020) 55(1):123–8. doi: 10.1080/00365521.2019.1707275

3. Sha J, Dong Y, Niu H. A prospective study of risk factors for in-hospital mortality in patients with malignant obstructive jaundice undergoing percutaneous biliary drainage. *Medicine (Baltimore)*. (2019) 98(15):e15131. doi: 10.1097/MD.0000000000015131

4. Varadarajulu S. Endoscopic ultrasound-guided biliary drainage for palliation of malignant obstructive jaundice. *Gastroenterol Hepatol*. (2019) 15(2):105–7. doi: 10.2147/OTT.S312162

5. Li Z, Jiao D, Han X, Liu Z. A comparative study of self-expandable metallic stent combined with double (125)I seeds strands or single (125)I seeds strand in the treatment of advanced perihilar cholangiocarcinoma with malignant obstructive jaundice. *Onco Targets Ther*. (2021) 14:4077–86. doi: 10.2147/OTT.S312162

6. Wu JZ, Li CL, Shi HB, Liu S, Yang W, Zhou WZ. Hepatic arterial infusion chemotherapy following simultaneous metallic stent placement and iodine-125 seed strands for advanced cholangiocarcinoma causing malignant obstructive jaundice: a propensity score matching study. *Jpn J Radiol*. (2022) 40(4):396–403. doi: 10.1007/s11604-021-01212-7

7. Yao J, Kong Y, Wang C, Wei Y, Li H, Liu C. Endobiliary ablation combined with immune nutrition improves quality of life: a preliminary clinical study in patients with advanced malignant obstructive jaundice. *Med Sci Monit*. (2022) 28:e936863. doi: 10.12659/MSM.936863

8. Villegas L, Jones D, Lindberg G, Chang C, Tesfay S, Flemin JB. Laparoscopic choledochojejunostomy via PTFE-covered stent successfully achieves internal drainage of common bile duct obstruction. *HPB (Oxford)*. (2005) 7(2):149–54. doi: 10.1080/13651820410016723

9. Rizzo A, Ricci AD, Frega G, Palloni A, De Lorenzo S, Abbati F, et al. How to choose between percutaneous transhepatic and endoscopic biliary drainage in malignant obstructive jaundice: an updated systematic review and meta-analysis. *In Vivo*. (2020) 34(4):1701–14. doi: 10.21873/invivo.11964

10. Tang K, Sui L-L, Xu G, Zhang T, Liu Q, Liu X-F. Effects of different palliative jaundice reducing methods on immunologic functions in patients with advanced malignant obstructive jaundice. *Anticancer Res*. (2017) 37(8):4665–70. doi: 10.21873/anticanres.11870

11. Duan F, Cui L, Bai Y, Li X, Yan J, Liu X. Comparison of efficacy and complications of endoscopic and percutaneous biliary drainage in malignant obstructive jaundice: a systematic review and meta-analysis. *Cancer Imaging*. (2017) 17(1):27. doi: 10.1186/s40644-017-0129-1

12. Wu J, Song L, Zhang Y, Zhao DY, Guo B, Liu J. Efficacy of percutaneous transhepatic cholangiodrainage (PTCD) in patients with unresectable pancreatic cancer. *Tumour Biol*. (2014) 35(3):2753–7. doi: 10.1007/s13277-013-1363-1

13. Zhu L, Chen X. Change and significance of T-cell subsets and TNF-alpha in patients with advanced malignant obstructive jaundice treated by percutaneous transhepatic biliary external and internal drainage. *Front Med China*. (2007) 1 (4):364–8. doi: 10.1007/s11684-007-0070-y

14. Hori T, Aisu Y, Yamamoto M, Yasukawa D, Iida T, Yagi S, et al. Laparoscopic approach for choledochojejunostomy. *Hepatobiliary Pancreat Dis Int*. (2019) 18(3):285–8. doi: 10.1016/j.hbpd.2019.04.004

15. Lee JS, Hong TH. Laparoscopic choledochojejunostomy in various hepatobiliary and pancreatic surgeries: a single surgeon's experience. *J Laparoendosc Adv Surg Tech A*. (2015) 25(4):305–10. doi: 10.1089/lap.2014.0539

16. Kim H, Kwon H, Lim W, Moon BI, Paik NS. Quantitative assessment of the learning curve for robotic thyroid surgery. *J Clin Med*. (2019) 8(3):402. doi: 10.3390/jcm8030402

17. Cuendis-Velázquez A, Trejo-Ávila ME, Rosales-Castañeda E, Cardenas-Lailson E, Rojano-Rodríguez ME, Romero-Loera S, et al. Laparoscopic choledochoduodenostomy. *Cir Esp*. (2017) 95(7):397–402. doi: 10.1016/j.ciresp.2017.07.002

18. Kays JK, Koniaris LG, Milgrom DP, Nakeeb A. Biliary bypass with laparoscopic choledochoduodenostomy. *J Gastrointest Surg*. (2018) 22 (5):928–33. doi: 10.1007/s11605-017-3663-z

19. Senthilnathan P, Sharma D, Sabnis SC, Srivatsan Gurumurthy S, Senthil Anand E, Nalankilli VP, et al. Laparoscopic choledochoduodenostomy as a reliable rescue procedure for complicated bile duct stones. *Surg Endosc*. (2018) 32(4):1828–33. doi: 10.1007/s00464-017-5868-3

20. Rhodes M, Nathanson L. Laparoscopic choledochoduodenostomy. *Surg Laparosc Endosc*. (1996) 6(4):318–21. doi: 10.1097/00019509-199608000-00015

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# Impact of sarcopenia on postoperative pulmonary complications after gastric cancer surgery: A retrospective cohort study

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**Background:** Few studies have investigated the relationship between sarcopenia and postoperative pulmonary complications (PPCs) after gastric cancer surgery. This study aimed to explore the impact of sarcopenia on PPCs in patients who had undergone gastric cancer surgery.

**Methods:** We included patients who underwent a transabdominal radical gastrectomy between June 2016 and October 2020. Patients were divided into two groups according to the median prevalence rate of lumbar triplane skeletal muscle index (L3 SMI): sarcopenia group (≤37.5% percentile in male and female group) and non-sarcopenia group (>37.5% percentile in male and female group). Baseline characteristics, intraoperative and postoperative conditions, pulmonary complications, and overall complications were compared between the two groups. The primary outcome was the incidence of PPCs. The secondary outcomes were overall postoperative complications and length of stay (LOS).

**Results:** Among the 143 patients included, 50 had sarcopenia and 93 had not. Compared to the non-sarcopenia group, the sarcopenia group had a higher the incidence of PPCs (22.0% vs. 8.6%, $P = 0.024$). The incidence of overall postoperative complications in the sarcopenia group was higher than that in the non-sarcopenia group (36.00% vs. 20.43%, $P = 0.043$). There was no significant difference in the LOS between the two groups.

**Conclusions:** Our research indicates that sarcopenia, preoperative comorbidities, and longer duration of intraoperative oxygen saturation <95% were risk factors for PPCs. Sarcopenia is an independent risk factor for postoperative complications. Given that our results provided a correlation rather than causation, future prospective randomized trials are needed to confirm the relationship between sarcopenia and prognosis.

KEYWORDS

sarcopenia, skeletal muscle index (SMI), anesthesia, postoperative pulmonary complications (PPCs), gastric cancer

# Introduction

Sarcopenia is a complex age-related syndrome characterized by progressive and generalized loss of skeletal muscle mass and function (1, 2), with the potential for physical disability, loss of independence, and adverse consequences, such as death (3, 4). The etiology of sarcopenia may be related to skeletal muscle disuse, endocrine changes, chronic wasting disease, systemic inflammatory responses, insulin resistance, and malnutrition (5, 6). Excessive inflammatory responses and chronic wasting disease largely contribute to sarcopenia, especially in patients with cancer (7). Thus, sarcopenia and cancer are causally related. Patients with gastric cancer usually have a certain degree of anorexia and underlying metabolic changes such as increased energy consumption, catabolism, and inflammation. These direct effects are exacerbated by the combined effects of chemotherapy and major gastrectomy, resulting in decreased nutrient intake. Decreased nutritional intake in patients with gastric cancer can further aggravate the occurrence and development of sarcopenia (8, 9).

Gastric cancer is the fifth most common cancer and third leading cause of cancer-related deaths worldwide (10). Studies have shown that sarcopenia is an independent factor for postoperative complications and overall survival in patients with gastric cancer (11). Studies have shown that sarcopenia is very common in older people, with a prevalence of 5%–13% in people aged 60–70 years and 11%–50% in those aged >80 years of age. Large differences in prevalence are related to differences in the measurements and cutoffs used to define sarcopenia (12). The prevalence of sarcopenia among community residents in China was 4.8% among women and 13.2% among men aged ≥70 years (13). The prevalence of sarcopenia in patients with cancer has significantly increased by approximately 35.7% (14). With further aggravation of population aging, the number of older patients with gastric cancer will gradually increase (15). Surgery remains the most important treatment for gastric cancer (16). However, the high incidence of postoperative complications and low survival rate in such patients have always been a concern for clinicians (17, 18). Postoperative complications have been shown to affect overall survival (19). Predicting the risk of postoperative complications and how to better intervene in order to reduce postoperative complications have become the focus of attention. Previous studies have shown that patients with sarcopenia have a higher risk of postoperative complications, longer length of stay (LOS), and higher hospital costs than patients without sarcopenia (20). Among the postoperative complications, anastomotic leakage and pulmonary complications have the greatest influence on postoperative mortality and prolonged LOS (21). Anastomotic leakage has decreased with improvements in surgical techniques. Pneumonia or lung-related complications are the most common postoperative complications in individuals under 80 years of age (22). Accurately predicting the risk of complications and actively preventing and doing everything possible to reduce the occurrence of postoperative pulmonary complications (PPCs) have become the focus of surgeons and anesthesiologists. However, few studies have investigated the relationship between sarcopenia and PPCs after gastric cancer surgery. In this study, we aimed to investigate the impact of sarcopenia on PPCs in patients undergoing gastric cancer surgery and to identify other risk factors for post-operative pneumonia.

# Materials and methods

## Study design and patients

This single-center, retrospective cohort study used data obtained from the discharge medical records of patients undergoing gastrointestinal surgery from June 2016 to October 2020 in the Department of Gastrointestinal Surgery, West China Hospital, Sichuan University, Chengdu, China. This study was approved by the Biomedical Research Ethics Committee of West China Hospital, Sichuan University, and was registered at www.chictr.org.cn (ChiCTR1900026578).

## Inclusion and exclusion criteria

The inclusion criteria were as follows: age 18–75 years; American Society of Anesthesiologists (ASA) I–III, and plan to undergo transabdominal radical gastrectomy for clinical stage I-III gastric cancer. Patients were excluded if their medical records were incomplete or inaccurate, if they had missing abdominal computed tomography (CT), or if they had a history of radical gastric resection or preoperative chemotherapy. Proximal (PG), distal (DG), or total gastrectomy (TG) was performed by specialized surgeons, according to the Japanese Gastric Cancer Treatment Guidelines (23).

## Data collection

For each patient, the data were collected by trained surgeons, radiologists, and anesthesiologists. The surgeons were trained by experienced surgeons until they were sufficiently skilled and precise in data collection (as judged by an experienced surgeon).

The basic information was as follows: patient sociodemographic characteristics, clinical characteristics, surgical procedures, and outcomes. The intraoperative parameters examined were as follows: the types of resection, anesthesia method, operation time (min), mechanical

ventilation time (min), respiratory parameters (tidal volume [ml/kg], positive end expiratory pressure [PEEP], airway pressure, end-tidal carbon dioxide), circulation (systolic and diastolic blood pressure, heart rate, intraoperative vasoactive drug use), intraoperative pulse oximetry ($SPO_2$) < 95% duration, intraoperative infusion volume (colloid volume, crystalloid volume), urine volume, and medication data.

After surgery, we monitored: the time of removing the tracheal tube after surgery (min), days of hospitalization after surgery (days), hospitalization expenses (yuan), time of removing gastric tube after surgery (days), postoperative complications, and pulmonary complications within 15 days after surgery (24), postoperative destination, whether patient-controlled analgesia was used, postoperative pathological diagnosis, histological type, TNM stage, and readmission within 30 days of discharge.

For primary outcomes, we measured PPCs and for secondary outcome measures, we considered the severity classification of PPCs, other postoperative complications, and intra-abdominal infections.

## Diagnosis of sarcopenia

Sarcopenia was defined as low muscle mass, strength, and/or physical performance. Previous studies have shown that lumbar triplane skeletal muscle index (L3 SMI) on CT is the gold standard for estimating muscle quality (25). After professional training, imaging physicians identified and measured the muscle area of the L3 plane and divided it by the height squared ($m^2$) to obtain the skeletal muscle index of L3 SMI ($cm^2/m^2$) on the syngo Multimodality Workplace software (Siemens Medical Solutions, Forchheim, Germany).

Studies have shown that the median prevalence of sarcopenia in patients with gastric cancer is approximately 35.7% (14). Therefore, in this study, we used a median of 35.7% for grouping. In all the medical records collected, ≤ 35.7% of both sexes were classified as the sarcopenia group and >35.7% as the non-sarcopenia group. A data collection flowchart is presented in **Figure 1**.

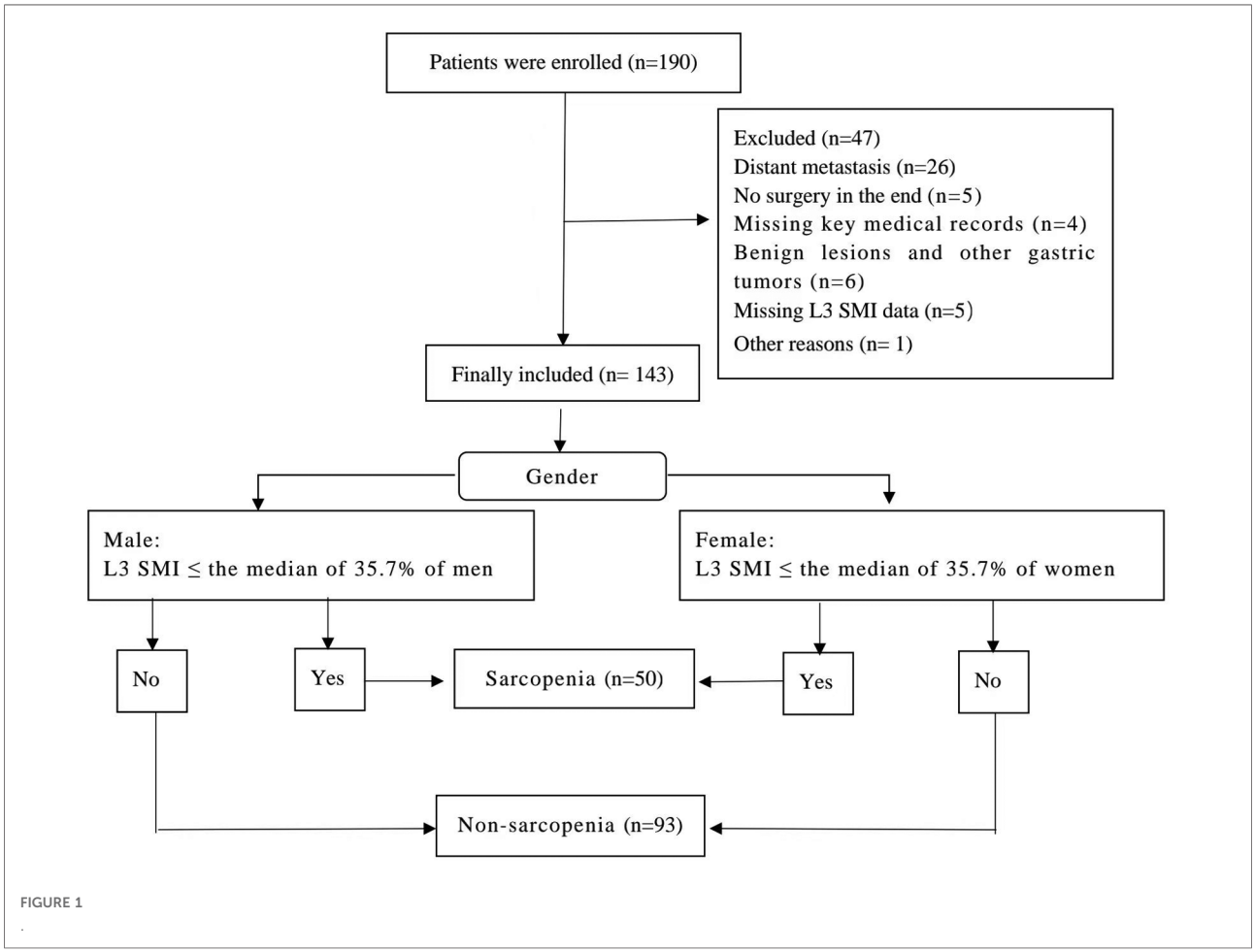


FIGURE 1

TABLE 1 Baseline characteristics of sarcopenia vs. non-sarcopenia groups.

| | Sarcopenia group ($n = 50$) | Non-sarcopenia group ($n = 93$) | $p$-value |
|---|---|---|---|
| Age (years) (Mean±SD) | 59.4 ± 10.1 | 55.9 ± 10.7 | 0.064 |
| Gender: Male (%) | 33 (66.0) | 62 (66.7) | 0.936 |
| Height (cm) M (P25, P75) | 163.5 (157.8, 170.0) | 165.0 (158.0, 170.0) | 0.726 |
| Weight (kg) M (P25, P75) | 53.3 (47.5, 60.3) | 63.0 (58.0, 70.0) | <0.001* |
| BMI (kg/m$^2$) M (P25, P75) | 20.0 (18.0, 21.7) | 23.2 (21.6,25.8) | <0.001* |
| Previous history of abdominal surgery (%) | 14/36 (28.0/72.0) | 19/74 (20.4/79.6) | 0.306 |
| Preoperative complication (%) | 20 (40.0) | 29 (31.2) | 0.289 |
| Hypertension (%) | 8 (16.0) | 12 (12.9) | 0.611 |
| Diabetes (%) | 4 (8.0) | 8 (8.6) | 1.000 |
| Chronic lung disease (%) | 7 (14.0) | 4 (4.3) | 0.081 |
| ASA grade (%) | | | |
| I | 0 (0) | 1 (1.1) | |
| II | 36 (72.0) | 81 (87.1) | |
| III | 14 (28.0) | 10 (10.8) | |
| $SPO_2 < 95\%$ when inhaling air (%) | 1 (2.0) | 6 (6.45) | 0.434 |
| Surgical approach (Billroth I/RYGB) (%) | 21/29 (42.0/58.0) | 28/65 (30.1/69.9) | 0.153 |
| Surgical types | | | 0.003* |
| Total gastrectomy | 21 (42.0) | 29 (31.2) | |
| Proximal gastrectomy | 0 (0) | 12 (12.9) | |
| Distal gastrectomy | 29 (58.0) | 52 (55.9) | |
| Pathological diagnosis adenocarcinoma (%) | 40 (80.0) | 89 (95.7) | 0.007* |
| Histologic Grade (%) | | | 0.783 |
| G2-G3 | 15 (30.0) | 30 (32.2) | |
| G2 | 9 (18.0) | 15 (16.1) | |
| G3 | 16 (32.0) | 38 (40.9) | |
| others | 10 (20.0) | 10 (10.8) | |
| TNM Stage | | | 0.143 |
| 1 | 11 (22.0) | 31 (35.5) | |
| 2 | 17 (34.0) | 20 (20.4) | |
| 3 | 22 (44.0) | 40 (43.0) | |
| Neutrophils ($10^9$/L) M (P25, P75) | 3.0 (2.4, 4.0) | 3.1 (2.5, 3.9) | 0.912 |
| Lymphocytes ($10^9$/L) M (P25, P75) | 1.6 (1.3, 1.9) | 1.6 (1.3, 1.9) | 0.741 |
| Mononuclear cell ($10^9$/L) M (P25, P75) | 0.4 (0.3, 0.5) | 0.4 (0.3, 0.5) | 0.599 |
| T-BIL (μmol/L) M (P25, P75) | 8.1 (6.4, 10.8) | 10.7 (8.3, 14.5) | 0.001* |
| ALT (IU/l) M (P25, P75) | 12.5 (9.0, 17.3) | 15.0 (12.0, 23.0) | 0.006* |
| AST (IU/L) M (P25, P75) | 19.0 (15.0, 22.3) | 19.0 (15.5, 23.0) | 0.529 |
| Albumin (g/L) Mean ± SD | 39.9 ± 4.0 | 42.3 ± 3.9 | 0.001* |
| Blood glucose (mmol/L) M (P25, P75) | 4.9 (4.6, 5.3) | 5.0 (4.6, 5.4) | 0.906 |
| Blood urea nitrogen (mmol/L) M (P25, P75) | 4.7 (4.0, 6.0) | 4.80 (4.4, 5.5) | 0.588 |
| Serum creatinine (μmol/L) Mean ± SD | 67.8 ± 15.3 | 67.9 ± 13.8 | 0.963 |
| eGFR (ml/min/1.73 m$^2$) M (P25, P75) | 98.6 (88.7, 104.3) | 96.7 (90.5, 104.3) | 0.943 |
| Triglyceride (mmol/L) M (P25, P75) | 1.1 (0.9, 1.5) | 1.3 (0.8, 1.7) | 0.175 |
| Total cholesterol (mmol/L) Mean ± SD | 4.3 ± 0.8 | 4.4 ± 0.9 | 0.396 |
| HDL (mmol/L) M (P25, P75) | 1.2 (1.1, 1.5) | 1.2 (0.9, 1.4) | 0.369 |
| LDL (mmol/L) Mean ± SD | 2.5 ± 0.7 | 2.6 ± 0.7 | 0.348 |
| LDH (IU/L) Mean ± SD | 146.1 ± 28.3 | 155.6 ± 25.8 | 0.056 |
| Transferrin (g/L) M (P25, P75) | 2.2 (2.0, 2.5) | 2.2 (2.0, 2.7) | 0.563 |

(*continued*)

TABLE 1 Continued

| | Sarcopenia group ($n = 50$) | Non-sarcopenia group ($n = 93$) | $p$-value |
|---|---|---|---|
| Prealbumin (mg/L) M (P25, P75) | 204.0 (174.0, 229.0) | 222.5 (184.3, 257.5) | 0.029* |
| AFP (ng/ml) M (P25, P75) | 2.5 (1.8, 4.0) | 3.0 (2.2, 4.1) | 0.183 |
| CEA (ng/ml) M (P25, P75) | 2.0 (1.3, 3.7) | 2.0 (1.1, 3.2) | 0.584 |
| CA19-9 (U/ml) M (P25, P75) | 10.0 (5.8, 27.4) | 10.9 (7.2, 15.2) | 0.835 |
| CA-125 (U/ml) M (P25, P75) | 14.1 (8.4, 20.1) | 12.2 (9.4, 17.1) | 0.723 |

SD, standard deviation; M, median; P25, 25% quantile; P75, 75% quantile; BMI, body mass index; ASA, american society of anesthesiologists classification; $SPO_2$, pulse oximetry; RYGB, roux-en-Y gastric bypass; TNM, stage tumor-lymph node-metastasis staging; T-BIL, total bilirubin; ALT, alanine aminotransferase; AST, aspartate aminotransferase; eGFR, estimated glomerular filtration rate; HDL, high density lipoprotein; LDL, low-density lipoprotein; LDH, lactate dehydrogenase; AFP, alpha-fetoprotein; CEA, carcinoembryonic antigen; CA19-9, carbohydrate antigen 19-9; CA-125, carbohydrate antigen 125.
*statistically significant ($P < 0.05$).

## Complications definition

PPCs were defined as any of the following postoperative conditions within 15 days after operation: initial ventilation support for >48 h, re-intubation due to respiratory failure or pneumonia, respiratory infection, respiratory failure, bronchospasm, atelectasis, pleural effusion, pneumothorax, or aspiration pneumonia (26). The severity of PPCs was classified as 0–5, in which 0 indicates that PPCs have no symptoms or signals, 1–4 indicates gradual deterioration of complications, and 5 indicates death before discharge (27). A grade of at least 2 was defined as severe PPCs. Postoperative complications were defined as any deviation from the normal postoperative course and were graded according to the Clavien-Dindo classification (28).

## Statistical analysis

The Shapiro–Wilk test was used to test the normality of continuous variables. The Student's *t*-test was used for quantitative data of normal and approximately normal distribution, and the mean±standard deviation was used for quantitative data of severely skewed distributions. For such distributions, the rank sum test was used and the data were expressed as median (25% quantile, 75% quantile). Categorical variables were analyzed using the chi-squared or Fisher's exact test, and classified variable data were expressed as numbers and percentages. Univariate logistic regression analysis was used for the univariate analysis. Variables with significant trends and known prognostic values, such as age, were selected as potential parameters in the univariate analysis. Forward stepwise variable selection was used to establish a multivariate logistic regression. All tests were bilateral (except for logistic regression analysis) and were considered statistically significant at $P < 0.05$. The IBM SPSS statistical software version 23.0 (SPSS Inc., Chicago, IL) was used for the statistical analysis.

# Results

## Comparison of baseline characteristics

A total of 143 patients met the inclusion criteria and were included in the study. The basic patient information is summarized in **Tables 1**, **2**. They were divided into sarcopenia and non-sarcopenia groups according to the abdominal CT-guided L3 SMI, and the median prevalence rate was 35.7%.

Compared with those in the non-sarcopenia group, the patients in the sarcopenia group had lower body weight (kg) ($P < 0.001$), BMI (kg/m$^2$) ($P < 0.001$), and preoperative blood albumin ($P = 0.001$). In addition, serum prealbumin (mg/L) ($P = 0.029$), T-BIL (μM) ($P = 0.001$), and ALT (IU/L) ($P = 0.006$) levels were also lower. The intraoperative small dose of remifentanil (μg/h) was lower ($P = 0.041$). The intraoperative tidal volume (ml/kg) of kilogram body weight (ml/kg) in the sarcopenia group was larger than that in the non-sarcopenia group ($P < 0.001$). As for the types of surgical resection, most distal gastrectomies were performed in the two groups, accounting for >50% in each group, while there was no proximal gastrectomy in the sarcopenia group ($P = 0.003$). The postoperative use of an intravenous analgesia pump was lower in the sarcopenia group than in the non-sarcopenia group ($P = 0.037$). In terms of pathological diagnosis, adenocarcinoma was lower in the sarcopenia group than in the non-sarcopenia group ($P = 0.007$). Other preoperative and intraoperative factors were not significantly different between the two groups.

## Comparison of short-term outcomes

Compared with the non-sarcopenia group, the sarcopenia group had worse outcomes for the incidence of the PPCs ($P = 0.024$). Moreover, the incidence of postoperative

TABLE 2 Perioperative management of sarcopenia vs. non-sarcopenia groups.

| | Sarcopenia group ($n = 50$) | non-sarcopenia group ($n = 93$) | $p$-value |
|---|---|---|---|
| Operation time (mins) M (P25, P75) | 154.0 (130.0, 177.5) | 150.0 (130.0, 179.3) | 0.957 |
| Colloidal fluid (ml/h) Mean±SD | 203.0 ± 129.5 | 215.0 ± 97.9 | 0.536 |
| Crystal liquid (ml/h) Mean±SD | 555.6 ± 166.2 | 540.1 ± 165.6 | 0.595 |
| Urine volume (ml/h/kg) M (P25, P75) | 1.5 (0.5, 2.6) | 1.3 (0.8,2.1) | 0.971 |
| Sevoflurane (ml/h) M (P25, P75) | 15.44 (0, 20.9) | 15.4 (0, 19.6) | 0.662 |
| Desflurane (ml/h) M (P25, P75) | 0 (0, 3.9) | 0 (0, 6.6) | 0.742 |
| Propofol (mg/h) M (P25, P75) | 155.9 (32.2, 256.1) | 63.6 (32.2, 324.4) | 0.563 |
| Dexmedetomidine (ug/h) M (P25, P75) | 5.9 (0, 18.2) | 14.2 (0, 22.0) | 0.065 |
| Sufentanil (ug/h) M (P25, P75) | 13.2 (11.4, 15.6) | 14.9 (11.8, 16.5) | 0.211 |
| Remifentanil (ug/h) Mean±SD | 377.0 ± 156.7 | 446.6 ± 208.0 | 0.041* |
| Cisatracurium (mg/h) Mean±SD | 8.2 ± 1.8 | 8.7 ± 2.5 | 0.181 |
| Use of higher doses of vasoactive drugs (%) | 12 (24.0) | 17 (18.3) | 0.417 |
| Use of high-dose antihypertensive drugs (%) | 3 (6.0) | 4 (4.3) | 0.966 |
| Use higher doses of vasopressors (%) | 9 (18.0) | 14 (15.1) | 0.647 |
| Intraoperative heat preservation (%) | 12 (24.0) | 18 (19.35) | 0.536 |
| Intraoperative blood transfusion (%) | 5 (10.0) | 4 (4.3) | 0.329 |
| Duration of intraoperative $SPO_2 < 95\%$ (mins) M (P25, P75) | 0 (0, 0) | 0 (0, 0) | 0.457 |
| Ventilation strategy Capacitance Control/Voltage Control (%) | 28/19 (56.0/38.0) | 58/26 (62.4/28.0) | 0.465 |
| Mechanical ventilation time (mins) Mean±SD | 201.1 ± 35.7 | 202.8 ± 40.2 | 0.808 |
| Mean of peep ($cmH_2O$) M (P25, P75) | 2.9 (2, 3) | 2.8 (2, 3) | 0.950 |
| Peak airway pressure > 20 $cmH_2O$ $n$ (%) | 0 (0) | 4 (4.3) | 0.335 |
| Peak airway pressure > 15 cm $H_2O$ duration (mins) M (P25, P75) | 0 (0, 15) | 0 (0, 45) | 0.185 |
| Peak airway pressure > 15 $cmH_2O$ n (%) | 19 (38.0) | 42 (45.2) | 0.343 |
| Peak airway pressure > 15 $cmH_2O$ continues to exceed 30 min $n$ (%) | 9 (18.0) | 24 (25.8) | 0.262 |
| Peak airway pressure > 15 $cmH_2O$ continues to exceed 15 min $n$ (%) | 10 (20.0) | 30 (32.3) | 0.098 |
| Tidal volume (ml/kg) M (P25, P75) | 7.1 (6.3, 8.0) | 6.6 (5.8, 7.0) | <0.001* |
| $ETCO_2 > 45$ mmHg duration (mins) M (P25, P75) | 0 (0, 5) | 0 (0, 0) | 0.221 |
| Heart rate change ≥30% duration (mins) M (P25, P75) | 7.5 (0, 35.0) | 7.5 (0, 25.0) | 0.771 |
| Heart rate <55 beats/min duration (mins) M (P25, P75) | 10.0 (0,22.5) | 5.0 (0, 23.8) | 0.979 |
| Heart rate >100 beats/min duration (mins) M (P25, P75) | 0 (0,5.0) | 0 (0, 5.0) | 0.673 |
| SBP change ≥30% duration (mins) M (P25, P75) | 12.5 (0, 60.0) | 10.0 (1.3, 40.0) | 0.787 |
| SBP change ≥20% duration (mins) M (P25, P75) | 60.0 (27.5, 112.5) | 62.5 (31.3, 105.0) | 0.951 |
| DBP change ≥20% duration (mins) M (P25, P75) | 65.0 (30.0, 105.0) | 60.0 (26.3, 100.0) | 0.956 |
| DBP change ≥30% duration (mins) M (P25, P75) | 12.5 (0, 36.3) | 10.0 (5, 40.0) | 0.549 |
| Postoperative destination of the patient (ICU/ inpatient ward) (%) | 1/49 (2.0/98.0) | 1/92 (1.1/98.9) | |
| Postoperative analgesia pump (%) | 34 (68.0) | 76 (81.7) | 0.037* |

SD, standard deviation; M, median; P25, 25% quantile; P75, 75% quantile; PEEP, positive end expiratory pressure; $ETCO_2$, end-tidal carbon dioxide; SBP, systolic blood pressure; DBP, diastolic blood pressure; ICU intensive care unit.
*statistically significant ($P < 0.05$).

complications was higher in the sarcopenia group than in the non-sarcopenia group ($P = 0.043$). A total of three patients were then re-admitted within 30 days after discharge (one case in the sarcopenia group and two cases in the non-sarcopenia group). There were no significant differences in hospitalization cost, LOS, postoperative gastric tube extubation time, or postoperative endotracheal tube extubation time (Table 3).

## Risk factors of PPCs and postoperative complications

In the univariate analysis, age, sarcopenia, preoperative comorbidities, $SPO_2 < 95\%$ when inhaling air under air, and duration of intraoperative $SPO_2 < 95\%$ were risk factors for the PPCs. In the multivariate analysis that included these factors, sarcopenia (odds ratio [OR] 3.79, 95% confidence

TABLE 3 Outcomes of sarcopenia vs. non-sarcopenia groups.

| | Sarcopenia group ($n = 50$) | Non-sarcopenia group ($n = 93$) | *p*-value |
|---|---|---|---|
| Primary outcomes | | | |
| PPCs (%) | | | 0.024* |
| Grade 0 | 39 (78.0) | 85 (91.4) | |
| Grade 1 | 0 (0) | 2 (2.2) | |
| Grade 2 | 6 (12.0) | 3 (3.2) | |
| Grade 3 | 2 (4.0) | 2 (2.2) | |
| Grade 4 | 3 (6.0) | 1 (1.0) | |
| Grade 5 | 0 (0) | 0 (0) | |
| Severe PPCs (≥ Grade 2) | 11 (22.0) | 6 (6.5) | 0.009* |
| Secondary outcomes | | | |
| Postoperative complications (%) | | | 0.043* |
| Grade 0 | 32 (64.0) | 74 (79.6) | |
| Grade I | 7 (14.0) | 11 (11.8) | |
| Grade II | 5 (10.0) | 5 (5.4) | |
| Grade III | 3 (6.0) | 2 (2.2) | |
| Grade IV | 3 (6.0) | 1 (1.0) | |
| Grade V | 0 (0) | 0 (0) | |
| Abdominal infection *n* (%) | 4 (8.0) | 0 (0) | |
| Postoperative tracheal tube removal time (mins) M (P25, P75) | 12.5 (5.8, 25.3) | 10.0 (5.0, 20.0) | 0.150 |
| Postoperative gastric tube removal time (days) M (P25, P75) | 3.0 (0, 5.0) | 3.0 (0, 5.0) | 0.451 |
| Length of stay (days) M (P25, P75) | 7.0 (6.0, 8.0) | 7.0 (6.0, 8.0) | 0.684 |
| Number of readmissions within 30 days after discharge *n* (%) | 1 (2.0) | 2 (2.2) | |
| Hospital expenses (yuan) M (P25, P75) | 81,548.5 (77,895.8, 87,377.8) | 79,796.0 (72,411.0, 84,615.0) | 0.069 |

M, median; P25, 25% quantile; P75, 75% quantile; PPCs, postoperative pulmonary complications.
*statistically significant ($P < 0.05$).

interval [CI] 1.27–11.34, $P = 0.017$), preoperative comorbidities (OR 2.86, 95% CI 1.01–8.15, $P = 0.049$), and duration of intraoperative $SPO_2 < 95\%$ (OR 1.14, 95% CI 1.04–1.24. $P = 0.005$) may be risk factors for PPCs. Univariate analysis also revealed that age, sarcopenia, preoperative comorbidities, and the duration of intraoperative $SPO_2 < 95\%$ were risk factors for severe PPCs. Multivariate regression analysis showed that sarcopenia (OR 5.10, 95% CI 1.63–16.00, $P = 0.005$) and the duration of intraoperative $SPO_2 < 95\%$ (OR 1.12; 95% CI 1.02–1.22, $P = 0.016$) were independent risk factors for severe PPCs in patients after gastric cancer surgery (Table 4).

Univariate analyses also found that sarcopenia was the risk factor for postoperative complications. Multivariate regression analysis with age using binary logistic regression showed that sarcopenia (OR 2.19, 95% CI 1.02–4.72, $P = 0.045$) was an independent risk factor for postoperative complications after gastric cancer surgery (Table 5).

## Discussion

We performed a single-center cohort study to investigate the effect of sarcopenia on PPCs in patients after gastric cancer surgery. We found that sarcopenia was associated with a higher incidence of postoperative complications and PPCs, compared to non-sarcopenia, in patients undergoing gastric cancer surgery. Multivariate regression analysis showed that sarcopenia, preoperative comorbidities, and the duration of intraoperative $SPO_2$ <95% were the risk factors for PPCs in patients undergoing radical gastrectomy. Sarcopenia and intraoperative $SPO_2$ <95% were still the risk factors for severe PPCs. In addition, sarcopenia was an independent risk factor for postoperative complications after gastrectomy.

Some studies have investigated the relationship between sarcopenia and postoperative complications following gastric cancer surgery (29–31). Patients with sarcopenia have a higher risk of postoperative complications and longer LOS. Zhou et al. indicated that sarcopenia is a strong independent risk factor for postoperative complications in older patients with gastric cancer (32). This is consistent with the results of our studies. The mechanism of sarcopenia leading to the increased risk of postoperative complications, especially pulmonary complications, remains unclear, and it is speculated that it may be related to the following possible mechanisms. Respiratory and swallowing muscles are affected by sarcopenia, which can lead to damage to the lungs and swallowing function. Impaired respiratory muscle function and swallowing function may lead to postoperative difficulty in expectoration, aspiration, postoperative pneumonia, and atelectasis (33, 34). Second, sarcopenia is associated with increased insulin resistance and increased circulation of proinflammatory cytokines, which may lead to the risk of postoperative acute lung injury (35). It has been reported that sarcopenia is associated with an increased inflammatory response to surgery (36). Increased inflammatory activity may also lead to pulmonary complications. Muscle fibers produce cytokines and other peptides such as interleukin-6, which affect the immune response by inhibiting the production of tumor necrosis factor-α and insulin resistance (37, 38). Sarcopenia may lead to immune senescence, which is characterized by impaired cellular immune function and increased inflammatory activity (39).

These factors may lead to PPCs. Fortunately, preoperative exercise through inspiratory muscle training, nutritional

TABLE 4 Univariate and multivariate analysis of factors associated with PPCs.

| | PPCs | | | | Severe PPCs | | | |
|---|---|---|---|---|---|---|---|---|
| | Univariate analysis | | Multi-factor analysis | | Univariate analysis | | Multi-factor analysis | |
| | OR (95% CI) | *p*-value | OR (95% CI) | *p*-value | OR (95% CI) | *p*-value | OR (95% CI) | *p*-value |
| Age (years) | 1.08 1.02–1.15) | 0.015* | | | 1.06 (1.00–1.13) | 0.047* | | |
| Sarcopenia vs. non-sarcopenia | 3.00 (1.12–8.04) | 0.029* | 3.79 (1.27–11.34) | 0.017* | 4.09 (1.41–11.85) | 0.009* | 5.10 (1.63–16.00) | 0.005* |
| Preoperative comorbidities | 3.11 (1.16–8.35) | 0.024* | 2.86 (1.01–8.15) | 0.049* | 3.19 (1.13–8.99) | 0.028* | | |
| $SPO_2$ < 95% when inhaling air | 5.58 (1.14–27.23) | 0.034* | | | 3.20 (0.57–17.97) | 0.186 | | |
| Duration of intraoperative $SPO_2$ < 95% | 1.11 (1.03–1.21) | 0.010* | 1.14 (1.04–1.24) | 0.005* | 1.09 (1.00–1.18) | 0.049* | 1.12 (1.02–1.22) | 0.016* |

PPCs postoperative pulmonary complications; $SPO_2$ pulse oximetry. Values in parentheses are percentages unless otherwise stated.
*statistically significant ($P < 0.05$).

TABLE 5 Univariate and multivariate analysis of factors associated with postoperative complications.

| | Univariate analysis | | Multi-factor analysis | |
|---|---|---|---|---|
| | OR (95% CI) | *p*-value | OR (95% CI) | *p*-value |
| Age (years) | 1.03 (0.99–1.08) | 0.098 | | |
| Sarcopenia vs. non-sarcopenia | 2.19 (1.02–4.72) | 0.045* | 2.19 (1.02–4.72) | 0.045* |

Values in parentheses are percentages unless otherwise stated. *statistically significant ($P < 0.05$).

support, and other preoperative interventions may improve muscle function in patients with sarcopenia and effectively reduce the incidence of PPCs (25, 40). Further prospective studies are required to verify this finding. Sarcopenia can be diagnosed using a questionnaire, action ability test, or L3 SMI on CT. In the future, more attention should be paid to the diagnosis of sarcopenia in different regions, races, and populations, and the relationship between sarcopenia and prognosis should be further explored.

Our study also showed that the longer the duration of $SPO_2$ < 95%, the higher the incidence of PPCs, especially the severe PPCs. Previous studies also found that a low $SPO_2$ was associated with increased mortality and mortality caused by pulmonary diseases (41). The duration of intraoperative $SPO_2$ < 95% may be related to the changes in pulmonary ventilation function caused by lung disease, mechanical ventilation lung injury, operation, and other inflammatory stimulations, which are currently recognized as indicators closely related to PPCs. This study indicated that the management of intraoperative mechanical ventilation could be further improved.

Interestingly, we found that tidal volume was larger in the sarcopenia group ($P < 0.001$) than in the non-sarcopenia group. In clinical practice, the tidal volume is often set according to the patient's body weight and pulmonary function. Compared to the non-sarcopenia group, the sarcopenia group had a smaller body weight but a larger tidal volume. Studies have shown that mechanical ventilation itself can induce inflammation and cooperate with surgery-induced responses. This magnifying inflammatory cascade reaction leads to lung injury and systemic multiple-organ failure.

Sarcopenia negatively affects the prognosis of patients who require mechanical ventilation, increasing all-cause mortality in these patients (42). This may be related to nutritional status, chronic inflammatory reaction, changes in hormone levels, and lack of physical activity in sarcopenia. Some studies have shown that low tidal volume can reduce pulmonary and systemic inflammatory responses compared with conventional tidal volume (43, 44). Mechanical ventilation with a high tidal volume may cause injury in healthy lungs (45, 46). Although the tidal volume of the two groups in this study did not exceed 10 ml/kg, it was higher in the sarcopenia group than in the non-sarcopenia group. There was no significant difference in the average value of PEEP between the two groups (the average value was approximately 2.8–2.9). Some studies have shown that the use of lower levels of PEEP may make the small airways open and close repeatedly, resulting in atelectasis and accelerating the development of pulmonary complications (47, 48). Multifaceted lung-protective ventilation strategies for high-risk patients, combined with low tidal volume, reopening of collapsed alveoli, and moderate levels of PEEP, can prevent further collapse (49). This would help reduce the incidence of postoperative atelectasis, improve clinical results, and reduce the consumption of medical resources. Whether the existing lung-protective ventilation strategy is the best perioperative ventilation management mode for patients with sarcopenia,

and how to individualize PEEP and tidal volume to reduce the increase in PPCs caused by mechanical ventilation remain open questions.

Given the adverse effects of sarcopenia on mortality and hospital outcomes, sarcopenia is often considered a treatable indicator in adult respiratory medical treatment (50). It also shows that the prognosis of patients with sarcopenia can be improved through clinical intervention. Previous studies have shown that rehabilitation exercises, nutritional support, and growth hormone supplementation can improve the muscle mass and prognosis of patients with mechanical ventilation (51–53). However, to date, accurate intervention for sarcopenia has been the focus of attention in patients with oligomyopathy. These findings suggest that doctors should pay more attention to the perioperative respiratory system, intraoperative ventilation management, and postoperative lung rehabilitation. It is not limited to the preoperative evaluation and intraoperative management of anesthesiologists, but also includes early identification and intervention by surgeons, rehabilitation doctors, and nurses to reduce its effect on the poor prognosis of patients with sarcopenia. Therefore, it is necessary to carry out a unified standard diagnostic method, larger sample sizes, and multicenter prospective studies on sarcopenia intervention.

Our study bears several limitations. First, this was a single-center retrospective study with incomplete or, in limited cases, absent medical records, and a small sample size. The conclusions of this study need to be verified in additional multicenter prospective studies, involving larger samples. Second, the definitions of sarcopenia were different. In this study, CT-guided L3 SMI was directly used as an index to evaluate sarcopenia, but it was not diagnosed using a muscle strength test. Since this was a retrospective study, we could not comprehensively evaluate skeletal muscle function. Additionally, the cutoff value was not used in this study because of disease type and other factors. The cutoff value depends on measurement techniques, reference studies, and population availability. Moreover, the definition of sarcopenia is greatly influenced by race, population, sex, and other factors. Currently, considerable controversy remains. Therefore, we used the median prevalence rate of patients with gastric cancer to divide the patients into sarcopenia and non-sarcopenia groups. Because some patients visited the local hospital for revisit after surgery, the relevant data for a long time after surgery could not be accurately collected; therefore the long-term prognosis of the patients was not analyzed in this study.

## Conclusions

Our study demonstrates that the duration of intraoperative $SPO_2$ <95%, sarcopenia, and preoperative comorbidities were the risk factors for PPCs, especially severe PPCs. Furthermore, sarcopenia was an independent risk factor for postoperative complications. Future large randomized controlled trials and long-term follow-ups are needed to confirm the relationship between sarcopenia and prognosis.

## Data availability statement

The original contributions presented in the study are included in the article/Supplementary Material, further inquiries can be directed to the corresponding author/s.

## Ethics statement

The studies involving human participants were reviewed and approved by The Biomedical Research Ethics Committee of West China Hospital of Sichuan University. Written informed consent for participation was not required for this study in accordance with the national legislation and the institutional requirements.

## Author contributions

Conception and design: XZ, CD and XW; Provision of study materials or patients: XZ, CD and LH; Collection and assembly of data: XZ, CD and XW; Data analysis and interpretation: XZ, CD and RZ; Manuscript writing: XZ, CD and XW. All authors contributed to the article and approved the submitted version.

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## Conflict of interest

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

## Publisher's note

# References

1. Narici MV, Maffulli N. Sarcopenia: characteristics, mechanisms and functional significance. *Br Med Bull*. (2010) 95:139–59. doi: 10.1093/bmb/ldq008

2. Cruz-Jentoft AJ, Bahat G, Bauer J, Boirie Y, Bruyère O, Cederholm T, et al. Sarcopenia: revised European consensus on definition and diagnosis. *Age Ageing*. (2019) 48(4):601. doi: 10.1093/ageing/afz046

3. Lauretani F, Russo CR, Bandinelli S, Bartali B, Cavazzini C, Di Iorio A, et al. Age-Associated changes in skeletal muscles and their effect on mobility: an operational diagnosis of sarcopenia. *J Appl Physiol*. (2003) 95(5):1851–60. doi: 10.1152/japplphysiol.00246.2003

4. Cawthon PM, Marshall LM, Michael Y, Dam TT, Ensrud KE, Barrett-Connor E, et al. Frailty in older men: prevalence, progression, and relationship with mortality. *J Am Geriatr Soc*. (2007) 55(8):1216–23. doi: 10.1111/j.1532-5415.2007.01259.x

5. Douglas PJ, Short KR, Campbell WW, Elena V, Wolfe RR. Role of dietary protein in the sarcopenia of aging. *Am J Clin Nutr* (2008) (5):1562S. doi: 10.1093/ajcn/87.5.1562S

6. Sayer AA, Dennison EM, Syddall HE, Jameson K, Martin HJ, Cooper C. The developmental origins of sarcopenia: using peripheral quantitative computed tomography to assess muscle size in older people. *J Gerontol-Biol Sci Med Sci*. (2008) 63(8):835–40. doi: 10.1093/gerona/63.8.835

7. Dodson S, Baracos VE, Jatoi A, Evans WJ, Cella D, Dalton JT, et al. Muscle wasting in cancer cachexia: clinical implications, diagnosis, and emerging treatment strategies. *Annu Rev Med*. (2011) 62(1):265. doi: 10.1146/annurev-med-061509-131248

8. Stojcev Z, Matysiak K, Duszewski M, Banasiewicz T. The role of dietary nutrition in stomach cancer. *Contemp Oncol*. (2013) 17(4):343–5. doi: 10.5114/wo.2013.37213

9. Takiguchi S, Takata A, Murakami K, Miyazaki Y, Yanagimoto Y, Kurokawa Y, et al. Clinical application of ghrelin administration for gastric cancer patients undergoing gastrectomy. *Gastric Cancer*. (2014) 17(2):200. doi: 10.1007/s10120-013-0300-8

10. Siegel RL, Miller KD, Jemal A. Cancer statistics, 2016. *CA: Cancer J Clin*. (2016) 66(1):7–30. doi: 10.3322/caac.21332

11. Tamura T, Sakurai K, Nambara M, Miki Y, Toyokawa T, Kubo N, et al. Adverse effects of preoperative sarcopenia on postoperative complications of patients with gastric cancer. *Anticancer Res*. (2019) 39(2):987–92. doi: 10.21873/anticanres.13203

12. Morley JE. Sarcopenia: diagnosis and treatment. *J Nutr Health Aging*. (2008) 12(7):452–6. doi: 10.1007/BF02982705

13. Cheng Q, Zhu X, Zhang X, Li H, Du Y, Hong W, et al. A cross-sectional study of loss of muscle mass corresponding to sarcopenia in healthy Chinese men and women: reference values, prevalence, and association with bone mass. *J Bone Miner Metab*. (2014) 32(1):78–88. doi: 10.1007/s00774-013-0468-3

14. McGovern J, Dolan RD, Horgan PG, Laird BJ, McMillan DC. Computed tomography-defined low skeletal muscle Index and density in cancer patients: observations from a systematic review. *J Cachexia Sarcopenia Muscle*. (2021) 12(6):1408–17. doi: 10.1002/jcsm.12831

15. Kota K, Hiroko YS. Comparison of time trends in stomach cancer incidence (1973–2002) in Asia, from cancer incidence in five continents, vols iv–ix. *Jpn J Clin Oncol*. (2009) 39(1):71–2. doi: 10.1093/jjco/hyn150

16. Thrumurthy SG, Chaudry MA, Hochhauser D, Mughal M. The diagnosis and management of gastric cancer. *BMJ*. (2013) 347, f6367. doi: 10.1136/bmj.f6367

17. Takeshita H, Ichikawa D, Komatsu S, Kubota T, Okamoto K, Shiozaki A, et al. Surgical outcomes of gastrectomy for elderly patients with gastric cancer. *World J Surg*. (2013) 37(12):2891–8. doi: 10.1007/s00268-013-2210-7

18. Zhou CJ, Chen FF, Zhuang CL, Pang WY, Zhang FY, Huang DD, et al. Feasibility of radical gastrectomy for elderly patients with gastric cancer. *Eur J Surg Oncol*. (2016) 42(2):303–11. doi: 10.1016/j.ejso.2015.11.013

19. Yan-Mei B, Xin-Zu C, Cheng-Kun J, Ru-Bai Z, Yu-Fei G, Li-Bo Y, et al. Safety and survival benefit of surgical management for elderly gastric cancer patients. *Hepato-gastroenterol*. (2014) 61(134):1801–5.

20. Wang SL, Zhuang CL, Huang DD, Pang WY, Lou N, Chen FF, et al. Sarcopenia adversely impacts postoperative clinical outcomes following gastrectomy in patients with gastric cancer: a prospective study. *Ann Surg Oncol*. (2016) 23(2):556–64. doi: 10.1245/s10434-015-4887-3

21. Gertsen EC, Goense L, Brenkman HJF, van Hillegersberg R, Ruurda JP. Identification of the clinically most relevant postoperative complications after gastrectomy: a population-based cohort study. *Gastric Cancer*. (2020) 23(2):339–48. doi: 10.1007/s10120-019-00997-x

22. Wong JU, Tai FC, Huang CC. An examination of surgical and survival outcomes in the elderly (65-79 years of age) and the very elderly (≥80 years of age) who received surgery for gastric cancer. *Curr Med Res Opin*. (2020) 36(2):229–33. doi: 10.1080/03007995.2018.1520083

23. Suo J, Wei LI. Interpretation of Japanese gastric cancer association(jgca) gastric cancer treatment guidelines 2018-the 5th edition. *Chin J Pract Surg*. (2018) 24(1):1–21. doi: 10.1007/s10120-020-01042-y

24. Clavien PA, Barkun J, Oliveira MLD, Vauthey JN, Makuuchi M. The clavien-dindo classification of surgical complications: five-year experience. *Ann Surg*. (2009) 250(2):187. doi: 10.1097/SLA.0b013e3181b13ca2

25. Phillips SM. Nutritional supplements in support of resistance exercise to counter age-related sarcopenia. *Adv Nutr*. (2019) 6(4):452–60. doi: 10.3945/an.115.008367

26. Canet J, Gallart L, Gomar C, Paluzie G, Vallès J, Castillo J, et al. Prediction of postoperative pulmonary complications in a population-based surgical cohort. *Anesthesiol*. (2010) 113(6):1338. doi: 10.1097/ALN.0b013e3181fc6e0a

27. Hulzebos EH, Helders PJ, Favié NJ, De Bie RA, Brutel de la Riviere A, Van Meeteren NL. Preoperative intensive inspiratory muscle training to prevent postoperative pulmonary complications in high-risk patients undergoing cabg surgery: a randomized clinical trial. *Jama*. (2006) 296(15):1851–7. doi: 10.1001/jama.296.15.1851

28. Dindo D, Demartines N, Clavien PA. Classification of surgical complications: a new proposal with evaluation in a cohort of 6336 patients and results of a survey. *Ann Surg*. (2004) 240(2):205–13. doi: 10.1097/01.sla.0000133083.54934.ae

29. Yang Z, Zhou X, Ma B, Xing Y, Jiang X, Wang Z. Predictive value of preoperative sarcopenia in patients with gastric cancer: a meta-analysis and systematic review. *J Gastrointest Surg*. (2018) 22(11):1890–902. doi: 10.1007/s11605-018-3856-0

30. Kuwada K, Kuroda S, Kikuchi S, Yoshida R, Nishizaki M, Kagawa S, et al. Sarcopenia and comorbidity in gastric cancer surgery as a useful combined factor to predict eventual death from other causes. *Ann Surg Oncol*. (2018) 25(5):1160–6. doi: 10.1245/s10434-018-6354-4

31. Fukuda Y, Yamamoto K, Hirao M, Nishikawa K, Nagatsuma Y, Nakayama T, et al. Sarcopenia is associated with severe postoperative complications in elderly gastric cancer patients undergoing gastrectomy. *Gastric Cancer*. (2016) 19(3):986–93. doi: 10.1007/s10120-015-0546-4

32. Zhou CJ, Zhang FM, Zhang FY, Yu Z, Chen XL, Shen X, et al. Sarcopenia: a new predictor of postoperative complications for elderly gastric cancer patients who underwent radical gastrectomy. *J Surg Res*. (2017) 211:137. doi: 10.1016/j.jss.2016.12.014

33. Wakabayashi H, Sakuma K. Rehabilitation nutrition for sarcopenia with disability: a combination of both rehabilitation and nutrition care management. *J Cachexia Sarcopenia Muscle*. (2014) 5(4):269–77. doi: 10.1007/s13539-014-0162-x

34. Bahat G, Tufan A, Ozkaya H, Tufan F, Akpinar TS, Akin S, et al. in Male nursing home residents. *Aging Male*. (2014) 17(3):136–40. doi: 10.3109/13685538.2014.936001

35. Nishigori T, Okabe H, Tanaka E, Tsunoda S, Sakai Y. Sarcopenia as a predictor of pulmonary complications after esophagectomy for thoracic esophageal cancer. *J Surg Oncol*. (2016) 113(6):678–84. doi: 10.1002/jso.24214

36. Reisinger KW, Derikx JPM, Vugt JLAV, Meyenfeldt MFV, Hulsewé KW, Damink SWMO, et al. Sarcopenia is associated with an increased inflammatory response to surgery in colorectal cancer. *Clin Nutr*. (2016) 35(4):924–7. doi: 10.1016/j.clnu.2015.07.005

37. Pedersen BK, Febbraio MA. Muscles, exercise and obesity: skeletal muscle as a secretory organ. *Nat Rev Endocrinol*. (2012) 8(8):457–65. doi: 10.1038/nrendo.2012.49

38. Pedersen BK, Bruunsgaard H. Possible beneficial role of exercise in modulating low-grade inflammation in the elderly. *Scand J Med Sci Sports*. (2003) 13(1):56–62. doi: 10.1034/j.1600-0838.2003.20218.x

39. Bruunsgaard H, Pedersen BK. Effects of exercise on the immune system in the elderly population. *Immunol & Cell Biol*. (2000) 78(5):523–31. doi: 10.1046/j.1440-1711.2000.00965.x

40. N S. Preoperative inspiratory muscle training and postoperative complications. *J Am Med Assoc JAMA*. (2007) 297(7):697–9. doi: 10.1001/jama.297.7.697-a

41. Vold ML, Aasebø U, Wilsgaard T, Melbye H. Low oxygen saturation and mortality in an adult cohort: the tromsø study. *BMC Pulm Med*. (2015) 15:9. doi: 10.1186/s12890-015-0003-5

42. Jiang T, Lin T, Shu X, Song Q, Dai M, Zhao Y, et al. Prevalence and prognostic value of preexisting sarcopenia in patients with mechanical ventilation: a systematic review and meta-analysis. *Critical Care*. (2022) 26 (1):140. doi: 10.1186/s13054-022-04015-y

43. Michelet P, D'Journo XB, Roch A, Doddoli C, Marin V, Papazian L, et al. Protective ventilation influences systemic inflammation after esophagectomy: a randomized controlled study. *Anesthesiol*. (2006) 105(5):911–9. doi: 10.1097/00000542-200611000-00011

44. Determann RM, Royakkers A, Wolthuis EK, Vlaar AP, Choi G, Paulus F, et al. Ventilation with lower tidal volumes as compared with conventional tidal volumes for patients without acute lung injury: a preventive randomized controlled trial. *Critical Care*. (2010) 14(1):R1. doi: 10.1186/cc8230

45. Weingarten TN, Whalen FX, Warner DO, Gajic O, Schears GJ, Snyder MR, et al. Comparison of two ventilatory strategies in elderly patients undergoing Major abdominal surgery. *Br J Anaesth*. (2010) 104(1):16–22. doi: 10.1093/bja/aep319

46. Neto AS, Cardoso SO, Manetta JA, Pereira VGOM, Espósito DC, Manoela DOPP, et al. Association between use of lung-protective ventilation with lower tidal volumes and clinical outcomes among patients without acute respiratory distress syndromes. *Surv Anesthesiol*. (2014) 58(3):108–9. doi: 10.1097/01.SA.0000446366.05578.70

47. Benidixen HH, Hedley-Whyte J, Laver MB. Impaired oxygenation in surgical patients during general anesthesia with controlled ventilation. A concept of atelectasis. *Surv Anesthesiol*. (1964) 8(6):571. doi: 10.1056/NEJM196311072691901

48. Duggan M, Kavanagh BP. Pulmonary atelectasis: a pathogenic perioperative entity. *Anesthesiol*. (2005) 102(4):838–54. doi: 10.1097/00000542-200504000-00021

49. Futier E, Constantin JM, Pelosi P, Chanques G, Jaber S. Noninvasive ventilation and alveolar recruitment maneuver improve respiratory function during and after intubation of morbidly obese patients: a randomized controlled study. *Anesthesiol*. (2011) 114(6):1354–63. doi: 10.1097/ALN.0b013e31821811ba

50. McDonald VM, Osadnik CR, Gibson PG. Treatable traits in acute exacerbations of chronic airway diseases. *Chron Respir Dis*. (2019) 16:1479973119867954. doi: 10.1177/1479973119867954

51. Medrinal C, Combret Y, Prieur G, Robledo Quesada A, Bonnevie T, Gravier FE, et al. Comparison of exercise intensity during four early rehabilitation techniques in sedated and ventilated patients in icu: a randomised cross-over trial. *Critical Care*. (2018) 22(1):110. doi: 10.1186/s13054-018-2030-0

52. van Zanten ARH, De Waele E, Wischmeyer PE. Nutrition therapy and critical illness: practical guidance for the icu, post-icu, and long-term convalescence phases. *Critical Care*. (2019) 23(1):368. doi: 10.1186/s13054-019-2657-5

53. Hermans G, De Jonghe B, Bruyninckx F, Van den Berghe G. Interventions for preventing critical illness polyneuropathy and critical illness myopathy. *Cochrane Database Syst Rev*. (2009) 1:Cd006832. doi: 10.1002/14651858.CD006832.pub2

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# Novel end-to-side one-layer continuous pancreaticojejunostomy vs. end-to-end invaginated pancreaticojejunostomy in pancreatoduodenectomy: A single-center retrospective study

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**Background and Objective:** Postoperative pancreatic fistula (POPF) is the most common critical complication after pancreatoduodenectomy (PD) and is the primary reason for increased mortality and morbidity after PD. We aim to investigate the clinical significance of a novel approach, i.e., end-to-side one-layer continuous pancreaticojejunostomy, for patients with PD.
**Methods:** The clinical data of 65 patients who underwent pancreatoduodenectomy at the Xiangya Hospital, Central South University, from September 2020 to December 2021 were retrospectively analyzed.
**Results:** Forty patients underwent end-to-end invaginated pancreaticojejunostomy, and 25 underwent the novel end-to-side one-layer continuous pancreaticojejunostomy. No significant differences were observed in pancreatic fistula, intraperitoneal infection, intraperitoneal bleeding, reoperation, postoperative hospital stay, or perioperative death between the two groups. However, the novel end-to-side one-layer continuous pancreaticojejunostomy group had significantly shorter operation duration ($32.6 \pm 5.1$ min vs. $8.3 \pm 2.2$ min, $p < 0.001$). The incidence of pancreatic fistula in the novel pancreaticojejunostomy group was 12%, including two cases of grade A POPF and only one case of grade B POPF. No cases of grade C POPF occurred. No deaths were observed during the perioperative period.
**Conclusions:** The novel anastomosis method leads to a shorter operation duration than the traditional anastomosis method and does not increase postoperative complications. In conclusion, it is a simplified and feasible method for pancreatic anastomosis.

KEYWORDS

pancreaticoduodenectomy, pancreatic fistula, end-to-side pancreaticojejunostomy, one-layer continuous pancreaticojejunostomy, pancreatic anastomosis

# Introduction

Pancreaticoduodenectomy (PD) is a widely performed but challenging operation that involves multiple procedures to resect tumors in the periampullary region (pancreatic head and surrounding areas) (1, 2). The PD procedure has been a challenging operation since it was first performed in 1898 (3) and is characterized by high rates of perioperative mortality, morbidity, and postoperative complications (4, 5). In recent decades, efforts to optimize perioperative management, improve surgical techniques, and centralize pancreatic surgery care have reduced the postoperative mortality rate to less than 5%. However, the postoperative complication rate remains high, ranging from 40% to 50% (6–9).

The most common complication of pancreatoduodenectomy is postoperative pancreatic fistula (POPF), which has been shown to be one of the most intractable complications and can increase hospitalization costs and mortality (10, 11). Studies have shown that the occurrence of POPF is related to some important factors (12, 13), including the texture of the pancreas, blood supply to the tissues, the diameter of the main pancreatic duct (MPD), the quality of pancreaticojejunostomy (PJ), and the surgeon's experience; PJ is an independent risk factor for POPF (14).

It has been recognized that reconstruction after PD is technically challenging, and pancreaticojejunostomy methods and techniques are the main influential factors in pancreatic fistula. However, surgeons can improve the technical proficiency of pancreaticojejunostomy reconstruction by choosing a suitable anastomotic method and improving the quality of anastomosis (15). Therefore, a potential method of promoting surgeon proficiency in pancreatic anastomosis is to design a simplified and safe technique for this challenging reconstruction.

Biological healing is a novel concept of PJ that has been proposed by numerous surgeons in recent years (16–19). This novel theory emphasizes factors such as the blood supply of the tissues, the tension of the anastomotic stoma, healing of the pancreatic stump, and recovery of digestive function. "Wide, loose, and sparse" anastomosis has been recommended as a novel goal for PJ. Based on scholars Bassi and Miao's method (20, 21). With this novel theory of "biological healing," we developed a novel and innovative anastomotic method: end-to-side one-layer continuous pancreaticojejunostomy. Twenty-five patients have been treated with this novel method since 2020. As such, we conducted this single-center retrospective study to compare the clinical values and outcomes of PD patients undergoing end-to-end invaginated pancreaticojejunostomy with those undergoing the novel end-to-side one-layer continuous pancreaticojejunostomy.

# Materials and methods

## Patients and data

In this single-center retrospective trial, 65 patients with pathologically confirmed lesions in the pancreatic head and surrounding areas who underwent PD by either end-to-end invaginated pancreaticojejunostomy (Group A) or end-to-side one-layer continuous pancreaticojejunostomy (Group B) from September 2020 to December 2021 at the Department of General Surgery, Xiangya Hospital, Central South University, were enrolled.

The inclusion criteria were as follows: (1) adult patients (age from 18 to 80 years); (2) planned for selective pancreaticoduodenectomy; (3) no distant metastasis (including pelvic cavity, peritoneum, liver, lung, brain, bone, etc.) determined by ultrasound or CT; (4) not receiving radiotherapy and chemotherapy before surgery; (5) no history of other malignant tumors or associated with other organ dysfunction.

The exclusion criteria were as follows: (1) MPD could not be identified intraoperatively; (2) change to other surgical procedures, such as total pancreatectomy or segmental resection; (3) external drainage was added or occlusion of the MPD occurred for any reason; (4) resection combined with other organs; (5) pancreaticoduodenectomy combined with vascular resection and laparoscopic resection patients.

Clinical data, including baseline demographic characteristics, operation duration, and complications (including pancreatic fistula, intraperitoneal infection, intraperitoneal bleeding, reoperation, postoperative hospital stay, and perioperative death) were collected (**Tables 1**, **2**).

All operations were performed by the same highly experienced and qualified surgeon (more than 35 years of

TABLE 1 The general information of two groups.

| Variables | End-to-end sleeve anastomosis ($n = 40$) | End-to-side one-layer continuous anastomosis ($n = 25$) | *p*-value |
|---|---|---|---|
| Gender | | | 0.601 |
| Male | 24 | 15 | |
| Female | 16 | 10 | |
| Age (years) | 52.3 ± 11.4 | 55.1 ± 10.9 | 0.947 |
| Primary disease | | | 1.000 |
| Pancreatic head carcinoma | 13 | 8 | |
| Ampullary carcinoma | 16 | 10 | |
| Chronic pancreatitis | 2 | 1 | |
| Other | 9 | 6 | |

TABLE 2 Postoperative complications in two groups.

| Variables | End-to-end sleeve anastomosis ($n = 40$) | End-to-side one-layer continuous anastomosis ($n = 25$) | $p$-value |
|---|---|---|---|
| Pancreaticojejunostomy duration | 32.6 ± 5.1 min | 8.3 ± 2.2 min | <0.001 |
| Pancreatic fistula | 6 (15%) | 3 (12%) | 1.000 |
| Grade A | 3 (7.5%) | 2 (8%) | |
| Grade B | 2 (5%) | 1 (4%) | |
| Grade C | 1 (2.5%) | 0 (0%) | |
| Intraperitoneal infection | 1 (2.5%) | 0 (0%) | 1.000 |
| Intraperitoneal bleeding | 1 (2.5%) | 0 (0%) | 1.000 |
| Reoperation | 1 (2.5%) | 0 (0%) | 1.000 |
| Postoperative hospital stay (days) | 15.6 ± 6.1 | 14.8 ± 4.9 | 0.873 |
| Perioperative death | 1 (2.5%) | 0 (0%) | 1.000 |

clinical experience in pancreaticoduodenectomy) in the Department of General Surgery, Xiangya Hospital, Central South University. All work was reviewed and approved by the Ethics Committee of the Medical Council of Xiangya Hospital, Central South University. All patients or their legal representatives signed informed consent forms prior to surgery. According to Chinese law, this work was considered a quality-assured activity.

## Surgical procedure

All patients underwent pancreaticoduodenal resection. In accordance with the principle of radical cancer cure, we performed an operation to remove the entire tumor; clear the lymph node; skeletonize the hepatoduodenal ligament, the portal vein, and the superior mesenteric artery; and remove retroperitoneal tissue. The end-to-end pancreaticojejunostomy sleeve anastomosis used conventional child anastomosis. The method for the novel end-to-side one-layer continuous pancreaticojejunostomy was as follows. The surgeon made an all-layer continuous inverting suture between the pancreatic margin and the jejunum from the rear edge of the pancreas. Starting with a 2-0 Prolene slip line, the spacing was approximately 8–10 mm, and the margin was greater than 10 mm. Then, a support tube was built into the main pancreatic duct. When the rear wall was sutured, we placed the support tube into the jejunum. If the main pancreatic duct is greater than 4 mm in diameter, 2–3 stitches were sewed in the rear wall of the pancreatic duct and the posterior tissue together with the entire layer of jejunum. The front edge was turned from the rear edge, and the front edge of the pancreas and the other side of the jejunum were sewed with whole-layer suturing. The line was followed and knotted with the first line, and then pancreaticojejunostomy was completed. The critical points of our anastomotic technique included proper tension in the suture: not pulling too tightly in order to avoid pancreatic laceration, covering the entire pancreatic stump with the jejunal wall, not leaving dead space in between, and ensuring good contraposition of the opening of the jejunal wall (**Figure 1**).

## Postoperative management

All patients received routine medicine administration to prevent infection, suppress gastric acid, inhibit pancreatic secretion, protect liver function, support nutrition, and receive treatment for complications. Prophylactic octreotide was pumped continuously to all patients for 72 h after surgery. The amylase level of the drainage fluid was measured on postoperative days 1, 3, and 5 per the routine protocol and thereafter according to the surgeon's need.

## Postoperative complications

Postoperative complications of PD mainly include pancreatic fistula, postoperative intraperitoneal hemorrhage, anastomotic bleeding, biliary fistula, intestinal fistula, gastric emptying dysfunction, intraperitoneal infection, and so on. The diagnosis of pancreatic fistula standard adopts the International Team of Pancreatic Fistula (International Study Group of Pancreatic Fistula, ISGPF) definition of pancreatic fistula from 2005 (22): 3 days or more after surgery, amylase of drainage fluid from the drainage tube of surgical placement (or of subsequent percutaneous placement) is three times higher than the normal serum amylase limit. Patients with pancreatic fistula were divided into levels A, B, and C according to the clinical effect (**Supplementary Table S1**).

## Statistical analysis

The characteristics of the patients were summarized with frequencies and percentages (for categorical variables) or mean values ± standard deviations. SPSS 27.0 software (IBM Corporation, New York, United States) was used for data analysis. The measurement data were tested using the $t$-test, and the categorical data were tested by $\chi^2$ test. $p < 0.05$ indicated a significant difference.

# Results

Sixty-five patients who underwent PD were included from September 2020 to December 2021: 40 patients underwent

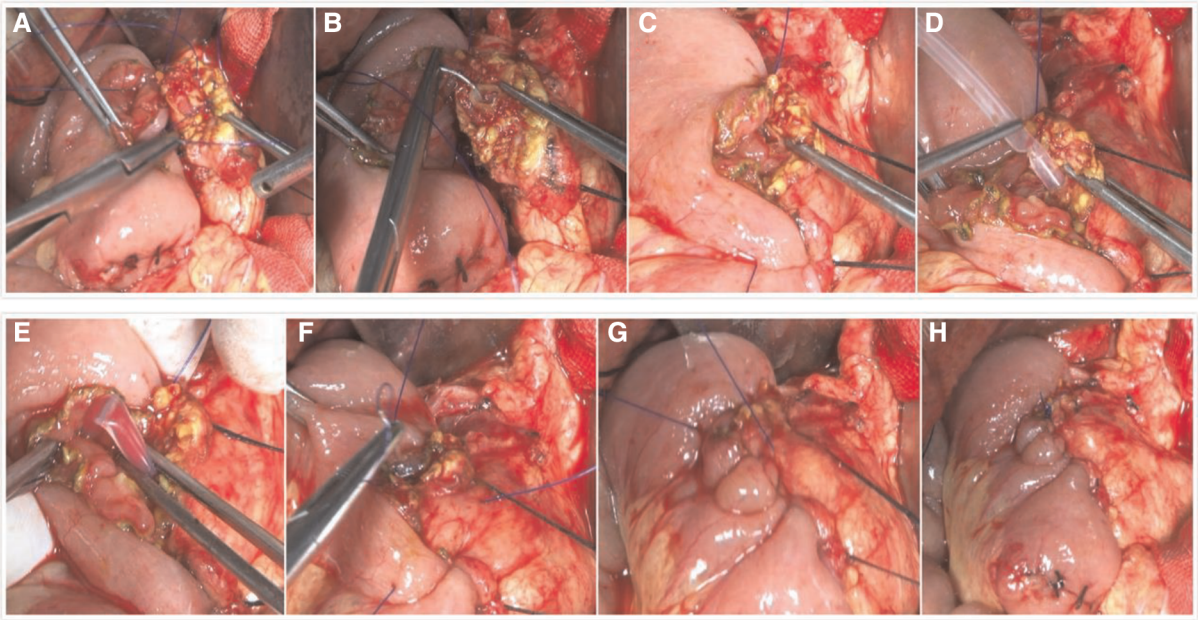


FIGURE 1
Intraoperative photographs of the novel end-to-side one-layer continuous pancreaticojejunostomy. (A) Perform all-layer continuous inverting suture between pancreatic margin and jejunal from the rear edge of the pancreas start with a 2-0 Prolene slip line. (B) Sew 2–3 stitches in the rear wall of the pancreatic duct and the posterior tissue together with the whole layer of the jejunum. (C) The suture of the rear wall is completed. (D) Build a support tube into the main pancreatic duct. (E) Put the support tube into the jejunum. (F) Turn to the front edge from the rear edge and sew the front edge of the pancreas and the other side of the jejunum with whole-layer suturing. (G) Take up the line. (H) Knot and complete pancreaticojejunostomy.

end-to-end invaginated pancreaticojejunostomy, and 25 underwent the novel end-to-side one-layer continuous pancreaticojejunostomy approach. There were no significant differences in age, sex, and primary disease of the patients between groups ($p > 0.05$). The baseline characteristics were also similar between groups (Table 1).

No significant difference was observed in the rates of pancreatic fistula, intraperitoneal infection, intraperitoneal bleeding, reoperation, postoperative hospital stay, and perioperative death. However, the pancreaticojejunostomy duration was significantly shorter ($8.3 \pm 2.2$ vs. $32.6 \pm 5.1$ min) in the novel end-to-side one-layer continuous pancreaticojejunostomy group than in the end-to-end invaginated pancreaticojejunostomy group ($p < 0.001$). The incidence of pancreatic fistula in the novel pancreaticojejunostomy was 12%, including two cases of grade A pancreatic fistula, one case of grade B pancreatic fistula, and no cases of grade C pancreatic fistula. No deaths occurred during the perioperative period (Table 2).

## Discussion

POPF is one of the most common and severe postoperative complications, and it can lead to a prolonged postoperative recovery time, intraperitoneal infection, intraperitoneal bleeding, and other complications. The onset of POPF can increase the mean length of hospital stay and medical costs, resulting in poor quality of life or even death (23, 24), which has been a main clinical challenge for pancreatic surgeons. Therefore, the healing of pancreatic-enteric anastomosis becomes very important for the prevention of pancreatic fistula.

Anastomosis between digestive organs, although under unique influences, such as digestive juice, motility, and tension, has a basic wound healing process, which can be divided into three periods (25): the inflammatory phase, proliferative phase, and remodeling phase. The inflammation phase usually occurs 0–7 days after surgery and presents mainly as local aggregation and infiltration of inflammatory cells. The proliferative neovascular response also performed relatively actively in this period. This period is prone to be accompanied by anastomotic leakage due to necrosis, bleeding, loss, and incomplete repair of the anastomotic tissue. The proliferative phase is generally 7–14 days after surgery. In this phase, inflammatory cells engulf necrotic tissue with significantly reduced leakage, an obvious proliferation of granulation tissue, an increasing number of fibroblasts, and large amounts of collagen fibers produced to repair wounds. In the remodeling phase, which occurs between 3 weeks and approximately 2 months after surgery, there is a further increase and gradually ordered collagen fibers that firm wound healing.

Pancreatic anastomotic healing has its specialty. First, the pancreas is a solid organ with slight toughness and can easily be torn when sutured. Second, anastomosis between the jejunum and pancreas, i.e., healing between different tissues, is influenced by pancreatic juice, bile, intestinal juice, and other types of digestive juice, which can be accompanied by severe necrosis and inflammatory exudation, long organization time, and slow epithelial regeneration. Therefore, anastomosis between the jejunum and pancreas has a markedly longer healing time than intestinal anastomosis, especially when the pancreas is soft with a small duct, which is a known risk factor for POPF.

Most clinicians believe that the novel anastomosis technique for pancreaticojejunostomy decreases the incidence of POPF in PD (26, 27). Therefore, surgeons have been concerned about exploring novel pancreaticojejunostomy techniques.

Since March 2020, we have performed a novel pancreaticojejunostomy method—pancreas–intestinal end-to-side one-layer continuous anastomosis—based on research related to full mouth whole-layer interrupted anastomosis that was conducted by Bassi, Miao ,and other scholars (20, 21).

Twenty-five patients underwent this novel pancreaticojejunostomy approach and achieved good results. No significant differences were observed in pancreatic fistula, intraperitoneal infection, intraperitoneal bleeding, reoperation, postoperative hospital stay, and perioperative death between the novel end-to-side one-layer continuous pancreaticojejunostomy group and the end-to-end invaginated pancreaticojejunostomy group.

Relevant studies have reported that the prevalence of POPF ranges from 10% to 40%, and a fistula rate of approximately 30% is generally accepted (28, 29). In our study, the incidence of PF in the novel pancreaticojejunostomy group was 12%, including two cases of grade A POPF, one case of grade B POPF, and no cases of grade C POPF. All patients with pancreatic fistula recovered after conservative treatment. There were no perioperative deaths.

Traditional pancreaticojejunostomy often attempts to reduce the occurrence of pancreatic fistula by using secure or even more stitched layers that are mechanically connected to the anastomosis. However, this anastomosis inhibits the natural biological healing process of pancreaticojejunostomy, so the incidence of postoperative pancreatic fistula after pancreaticojejunostomy does not decrease along with the various changes in the anastomosis method. Our understanding is that the goal of pancreaticojejunostomy is to establish pancreaticojejunostomy continuity between the pancreas and the jejunum by inducing biological healing (30). Stitching itself provides only the necessary conditions for the spatial proximity of a biological connection and the subsequent healing of organizations. A consistent and good approach should meet the following conditions: (1) anastomotic tissue has good blood supply; (2) margin involution is good; (3) damage to tissue cutting is minimal; and (4) it is simple and easy to operate. Thus, pancreaticojejunostomy could provide good conditions for healing.

The novel end-to-side one-layer continuous pancreaticojejunostomy group had significantly shorter pancreaticojejunostomy duration than the end-to-end invaginated pancreaticojejunostomy group. The novel method can be done within 6–10 min by experienced surgeons. In summary, this novel end-to-side one-layer continuous pancreaticojejunostomy is simpler and less time-consuming than the traditional method.

In clinical practice, we recognize that the end-to-side one-layer continuous pancreaticojejunostomy technique is superior with respect to simplicity and reliability. (1) Using Prolene slip lines (nonbiodegradable sutures that can maintain permanent tensile strength after being implanted into the tissue, extend with the creeping of the organization, and do not split due to fatigue) to suture a single layer continuously with an exact degree of sparse guarantees anastomosis and a better blood supply. This is a superior approach to try to do a tight suture with the line. (2) An appropriate degree of take-up and knotting, rather than cutting pancreatic tissue, can make the anastomosis margin moderately closer and keep the jejunum mucosa and pancreas margin neatly fit in the space, providing a good anatomic and physiological environment for healing. (3) Single-layer continuous sutures, rather than interrupted sutures, prevent tissue fragmentation due to repeated knotting (especially those with a soft pancreatic texture). The more complex the suture is, the more likely it is to affect the blood supply of the anastomosis, prolonging the phase of anastomotic inflammation and fiber decomposition and consequently leading to pancreatic fistula. Single continuous sutures shorten the phase of inflammation and fiber decomposition to make pancreaticojejunostomy heal faster with less scarring and reduced incidence of pancreatic fistula. (4) Single continuous sutures make the tension between the suture and anastomosis organizations distribute uniformly and softly so that the overall anti-tensile strength of anastomosis is high. (5) The intraductal support tube drains pancreatic secretin into the jejunum instead of accumulating in the anastomosis.

Our study has some limitations. First, as mentioned above, our sample size was relatively small. Second, this was a single-center, retrospective study because this novel pancreaticojejunostomy has been modified and is performed in our hospital currently. Therefore, multicenter randomized trials are needed for further research.

## Conclusions

In conclusion, we found that the novel end-to-side one-layer continuous pancreaticojejunostomy did not increase the

rate of postoperative complications after PD. However, the novel end-to-side one-layer continuous pancreaticojejunostomy showed advantages such as shorter pancreaticojejunostomy duration and a potentially reduced prevalence of pancreatic fistula. The findings need to be further validated with additional observational studies and animal experiments with large sample sizes. This novel method is feasible in both theory and practice. It is worthy of promotion and may bring significant clinical advantages to PD patients.

## Data availability statement

The original contributions presented in the study are included in the article/**Supplementary Material**, further inquiries can be directed to the corresponding authors.

## Ethics statement

The studies involving human participants were reviewed and approved by the Ethics Committee of the Medical Council of the Xiangya Hospital of Central South University. The patients/participants provided their written informed consent to participate in this study.

## Author contributions

DL and LJ: conceptualization, data curation, project administration. YL: conceptualization, project administration, analysis and interpretation of the data, validation. DL: funding acquisition, writing—original draft preparation. DL, LJ: analysis and interpretation of the data, formal analysis, visualization, investigation, software, investigation, methodology. LJ and XG: supervision, validation. All authors contributed to the article and approved the submitted version.

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## Conflict of interest

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

## Publisher's note



## Supplementary material

The Supplementary Material for this article can be found online at: https://www.frontiersin.org/articles/10.3389/fsurg.2022.980056/full#supplementary-material.

## References

1. Whipple AO, Parsons WB, Mullins CR. Treatment of carcinoma of the ampulla of vater. *Ann Surg.* (1935) 102:763–79. doi: 10.1097/00000658-193510000-00023

2. Gai YW, Wang HT, Tan XD. Pancreaticojejunostomy conducive to biological healing in minimally invasive pancreaticoduodenectomy. *J Gastrointest Surg.* (2022). doi: 10.1007/s11605-022-05339-4

3. Schnelldorfer T, Sarr MG. Alessandro codivilla and the first pancreatoduodenectomy. *Arch Surg.* (2009) 144(12):1179–84. doi: 10.1001/archsurg.2009.219

4. Kang CM, Lee JH. Pathophysiology after pancreaticoduodenectomy. *World J Gastroenterol.* (2015) 21(19):5794–804. doi: 10.3748/wjg.v21.i19.5794

5. Karim SAM, Abdulla KS, Abdulkarim QH, Rahim FH. The outcomes and complications of pancreaticoduodenectomy (Whipple procedure): cross sectional study. *Int J Surg.* (2018) 52:383–7. doi: 10.1016/j.ijsu.2018.01.041

6. McMillan MT, Allegrini V, Asbun HJ, Ball CG, Bassi C, Beane JD, et al. Incorporation of procedure-specific risk into the ACS-NSQIP surgical risk calculator improves the prediction of morbidity and mortality after pancreatoduodenectomy. *Ann Surg.* (2017) 265(5):978–86. doi: 10.1097/Sla.0000000000001796

7. Roberts KJ, Sutcliffe RP, Marudanayagam R, Hodson J, Isaac J, Muiesan P, et al. Scoring system to predict pancreatic fistula after pancreaticoduodenectomy: a UK multicenter study. *Ann Surg.* (2015) 261(6):1191–7. doi: 10.1097/Sla.0000000000000997

8. Nagai M, Sho M, Akahori T, Nishiwada S, Nakagawa K, Nakamura K, et al. Risk factors for late-onset gastrointestinal hemorrhage after pancreatoduodenectomy for pancreatic cancer. *World J Surg.* (2019) 43(2):626–33. doi: 10.1007/s00268-018-4791-7

9. Chen KT, Devarajan K, Hoffman JP. Morbidity among long-term survivors after pancreatoduodenectomy for pancreatic adenocarcinoma. *Ann Surg Oncol.* (2015) 22(4):1185–9. doi: 10.1245/s10434-014-3969-y

10. Bassi C, Marchegiani G, Dervenis C, Sarr M, Abu Hilal M, Adham M, et al. The 2016 update of the international study group (ISGPS) definition and grading of postoperative pancreatic fistula: 11 years after. *Surgery*. (2017) 161(3):584–91. doi: 10.1016/j.surg.2016.11.014

11. Deng Y, He S, Cheng Y, Cheng N, Gong J, Gong J, et al. Fibrin sealants for the prevention of postoperative pancreatic fistula following pancreatic surgery. *Cochrane Database Syst Rev*. (2020) 3:CD009621. doi: 10.1002/14651858.CD009621.pub4

12. Shrikhande SV, Sivasanker M, Vollmer CM, Friess H, Besselink MG, Fingerhut A, et al. Pancreatic anastomosis after pancreatoduodenectomy: a position statement by the International Study Group of Pancreatic Surgery (ISGPS). *Surgery*. (2017) 161(5):1221–34. doi: 10.1016/j.surg.2016.11.021

13. Casciani F, Trudeau MT, Asbun HJ, Ball CG, Bassi C, Behrman SW, et al. Surgeon experience contributes to improved outcomes in pancreatoduodenectomies at high risk for fistula development. *Surgery*. (2021) 169(4):708–20. doi: 10.1016/j.surg.2020.11.022

14. Soreide K, Healey AJ, Mole DJ, Parks RW. Pre-, peri- and post-operative factors for the development of pancreatic fistula after pancreatic surgery. *HPB (Oxford)*. (2019) 21(12):1621–31. doi: 10.1016/j.hpb.2019.06.004

15. Adams D. Why is a 0-fistula rate in pancreaticojejunostomy impossible? *Surgery*. (2021) 169(4):721–2. doi: 10.1016/j.surg.2020.12.002

16. Piao SJ, Pan ZJ, Qian CS, Jin XL. The effect of bilateral U-sutures in pancreaticojejunostomy in 75 consecutive cases. *Acta Chir Belg*. (2019) 119 (3):201–4. doi: 10.1080/00015458.2019.1610258

17. Zhou Y, Yang J, Wei L, Lin Q, Zheng S, Liu G, et al. A novel anastomosis technique facilitates pancreaticojejunostomy in total laparoscopic pancreaticoduodenectomy (with video). *Langenbecks Arch Surg*. (2021) 406 (8):2891–7. doi: 10.1007/s00423-021-02347-x

18. Tang W, Qiu JG, Li GZ, Zhao YF, Du CY. Clinical application of "double R" anastomosis technique in laparoscopic pancreaticoduodenectomy procedure. *Medicine (Baltimore)*. (2021) 100(21):e26204. doi: 10.1097/MD.0000000000026204

19. Miao Y, Dai C, Jiang K, Wu J, Gao W, Li Q, et al. Modified one-layer duct-to-mucosa pancreaticojejunostomy reduces pancreatic fistula after pancreaticoduodenectomy. *Pancreas*. (2015) 44(8):1397–8.

20. Bassi C, Falconi M, Molinari E, Mantovani W, Butturini G, Gumbs AA, et al. Duct-to-mucosa versus end-to-side pancreaticojejunostomy reconstruction after pancreaticoduodenectomy: results of a prospective randomized trial. *Surgery*. (2003) 134(5):766–71. doi: 10.1016/S0039-6060(03)00345-3

21. Miao YGWT, Jiang KR. Pancreatic-enteric anastomosis technique and pancreatic fistula. *J Hepatobiliary Surg*. (2009) 17(4):244–7.

22. Bassi C, Dervenis C, Butturini G, Fingerhut A, Yeo C, Izbicki J, et al. Postoperative pancreatic fistula: an international study group (ISGPF) definition. *Surgery*. (2005) 138(1):8–13. doi: 10.1016/j.surg.2005.05.001

23. Fuks D, Piessen G, Huet E, Tavernier M, Zerbib P, Michot F, et al. Life-threatening postoperative pancreatic fistula (grade C) after pancreaticoduodenectomy: incidence, prognosis, and risk factors. *Am J Surg*. (2009) 197(6):702–9. doi: 10.1016/j.amjsurg.2008.03.004

24. van Berge Henegouwen MI, De Wit LT, Van Gulik TM, Obertop H, Gouma DJ. Incidence, risk factors, and treatment of pancreatic leakage after pancreaticoduodenectomy: drainage versus resection of the pancreatic remnant. *J Am Coll Surgeons*. (1997) 185(1):18–24. doi: 10.1016/S1072-7515(97)00007-0

25. Wild T, Rahbarnia A, Kellner M, Sobotka L, Eberlein T. Basics in nutrition and wound healing. *Nutrition*. (2010) 26(9):862–6. doi: 10.1016/j.nut.2010.05.008

26. Berger AC, Howard TJ, Kennedy EP, Sauter PK, Bower-Cherry M, Dutkevitch S, et al. Does type of pancreaticojejunostomy after pancreaticoduodenectomy decrease rate of pancreatic fistula? A randomized, prospective, dual-institution trial. *J Am Coll Surgeons*. (2009) 208(5):738–47. doi: 10.1016/j.jamcollsurg.2008.12.031

27. Kawaida H, Kono H, Amemiya H, Hosomura N, Watanabe M, Saito R, et al. Anastomosis technique for pancreatojejunostomy and early removal of drainage tubes may reduce postoperative pancreatic fistula. *World J Surg Oncol*. (2020) 18(1):295. doi: 10.1186/s12957-020-02067-4

28. de Rooij T, van Hilst J, van Santvoort H, Boerma D, van den Boezem P, Daams F, et al. Minimally invasive versus open distal pancreatectomy (LEOPARD) A multicenter patient-blinded randomized controlled trial. *Ann Surg*. (2019) 269(1):2–9. doi: 10.1097/Sla.0000000000002979

29. Kleeff J, Diener MK, Z'graggen K, Hinz U, Wagner M, Bachmann J, et al. Distal pancreatectomy—risk factors for surgical failure in 302 consecutive cases. *Ann Surg*. (2007) 245(4):573–82. doi: 10.1097/01.sla.0000251438.43135.fb

30. Wei J, Liu X, Wu J, Xu W, Zhou J, Lu Z, et al. Modified one-layer duct-to-mucosa pancreaticojejunostomy reduces pancreatic fistula after pancreaticoduodenectomy. *Int Surg*. (2015).

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# Clinical outcomes of proximal gastrectomy with gastric tubular reconstruction and total gastrectomy for proximal gastric cancer: A matched cohort study

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**Background:** Proximal gastrectomy with gastric tubular reconstruction is a surgical procedure that can preserve function in patients with proximal gastric cancer. However, whether gastric tubular reconstruction with proximal gastrectomy has certain advantage in some aspects over total gastrectomy is controversial. To evaluate the benefit of gastric tubular reconstruction after proximal gastrectomy, we compared gastric tubular reconstruction with total gastrectomy for proximal gastric cancer.

**Method:** A total of 351 patients were enrolled. Concurrent total gastrectomy patients matched with the Proximal gastrectomy group in age, sex, body mass index, clinical stage, and ASA score were selected by propensity score matching. Preoperative basic information, perioperative indicators, histopathological features, postoperative complications and nutritional status, reflux were compared between the two groups.

**Results:** There was no significant difference in the incidence of reflux between two groups (14.8% and 6.5% respectively, $P = 0.085$). There were significant differences between the two groups in bowel function recovery ($2.29 \pm 1.16$ vs. $3.01 \pm 1.22$; $P = 0.039$) and start of soft diet ($4.06 \pm 1.81$ vs. $4.76 \pm 1.69$; $P = 0.047$). There were no significant differences between the two groups in nutritional status one year after surgery. However, the decrease in serum hemoglobin in the TG group at 3 and 6 months after surgery was significantly higher than that in the PG group ($P = 0.032$ and 0.046, respectively). One month after surgery, %BW loss in TG group was significantly lower than that in the PG group ($P = 0.024$).

**Conclusion:** The Proximal gastrectomy group has better clinical outcome and gastric tubular reconstruction is simple, similar complications and reflux rates, gastric tubular reconstruction may be more suitable for proximal gastric cancer.

KEYWORDS

proximal gastrectomy, gastric tubular reconstruction, reflux esophagitis, nutritional status, hemoglobin

## Introduction

The incidence of proximal gastric cancer is increasing worldwide (1–3). Proximal gastric cancer can occur in the upper 1/3 of the stomach or in the middle part of stomach; it constitutes more than 30% of gastric cancer cases. According to the Japan Gastric Cancer Association, the incidence of proximal gastric cancer increased by 0.8% from 2002 to 2011 (4). According to the 5th edition of the Japanese Guidelines for the Treatment of Gastric Cancer, total gastrectomy is the standard surgical treatment for upper 1/3 gastric cancer or AEG (5). The advantage of this surgical approach lies in the thorough dissection of lymph nodes that may metastasize and the avoidance of esophagogastric reflux complications. However, after total gastrectomy, patients will inevitably have nutritional metabolic disorders, especially patients with early proximal gastric cancer, which are more prominent (6–8). After total gastrectomy, the storage, mechanical grinding, secretion and other functions of the stomach are permanently lost, resulting in postoperative malnutrition, including decreased postoperative body mass, anemia, diarrhea, and dumping syndrome (8, 9).

Proximal gastrectomy of antireflux anatomical structures at the esophagogastric junction, including the cardia and His Angle, with simultaneous separation of the vagus nerve, resulted in an increased incidence of pyloric spasm and obstruction of residual stomach emptying (10). Some patients developed reflux esophagitis after surgery, which seriously affected the quality of life. Studies have shown that the incidence of postoperative reflux esophagitis is approximately 50% when traditional esophagogastrostomy is used in proximal gastrectomy (11, 12). To prevent RE, several reconstructive procedures after PG have been reported, such as double-flap (13), double-tract (14, 15), and jejunal interposition (16). However, these techniques are complicated, time-consuming and sometimes unsatisfactory.

Esophagogastrostomy was described by Shiraishi et al. in 1998 for the treatment of early proximal gastric cancer (17). This method excises part of the gastric antrum, reduces gastrin and gastric acid secretion, and objectively reduces reflux substances. After anastomosis, the tube stomach can make food pass through quickly and avoid food retention. The top of the residual stomach is similar to the fundus of the stomach, which can buffer the upward reflux of gastric juice and temporarily store the reflux of gastric juice. Therefore, the operation method has a good antireflux effect. Some research results show that compared with traditional residual gastroesophageal anastomosis, esophageal gastric tube anastomosis has a better quality of life for patients (18).

The purpose of this study was to determine whether gastric tubular reconstruction is a viable option after PG in terms of postoperative reflux and some nutritional indicators. We conducted a retrospective matched cohort study comparing the effects of gastric tubular reconstruction and total gastrectomy on patients with proximal gastric cancer.

## Methods

All patients with upper one-third gastric cancer consecutively received surgical treatment in the Gastrointestinal Surgery Department of Qingdao University Affiliated Hospital from January 2017 to February 2021. Upper third gastric cancer is defined as adenocarcinoma of the upper third of the stomach, with or without esophagogastric junction adenocarcinoma, according to the Classification of the Japan Gastric Cancer Association (JGCA) (19). The location of the primary carcinoma was determined by esophagogastroscopy. Inclusion criteria were that all patients had tumors located in the upper third of the stomach, clinical stage (CT1N0-1M0/CT2-3N0M0), R0 resection, and age 20–80. Exclusion criteria were neoadjuvant therapy, any malformation or ulcerative scarring of the distal stomach or duodenum, severe heart, lung, liver, kidney disease or mental abnormalities, and double primary carcinoma.

Patients were divided into two groups based on whether they underwent total or proximal gastrectomy. All patients underwent R0 resection. Patients underwent propensity score matching analysis, which adjusted for five factors, namely, age, sex, body mass index (BMI), pathological stage, and American Society of Anesthesiologists (ASA) body condition score, to offset selection bias. The matched whole stomach group was compared with the proximal stomach group based on demographic, clinical, surgical, and pathological features, postoperative outcomes (including early and late complications), postoperative nutritional status, and reflux esophagitis and reflux symptoms at endoscopic examination 1 year after surgery.

Preoperative tumor staging was assessed by computed tomography and gastroscopy. T stage and N stage were determined using the latest AJCC/UICC TNM staging system (20), and histological types were consistent with the Japanese classification of gastric cancer (19).

### Surgical procedure for PG with gastric tubular reconstruction and TG

All surgeries were performed by three upper gastrointestinal specialists using the same procedure (**Figures 1**, **2**).

Five ports were introduced, as shown in **Figure 1A**. Before reconstruction, lymph node dissection was completed according to the Japanese guidelines (5). Open the diaphragm angle, and bare the lower end of the esophagus by 2–3 cm. The esophagus was transected about the cardia with an endo GIA stapler. A 5 cm incision was made in the middle of the upper

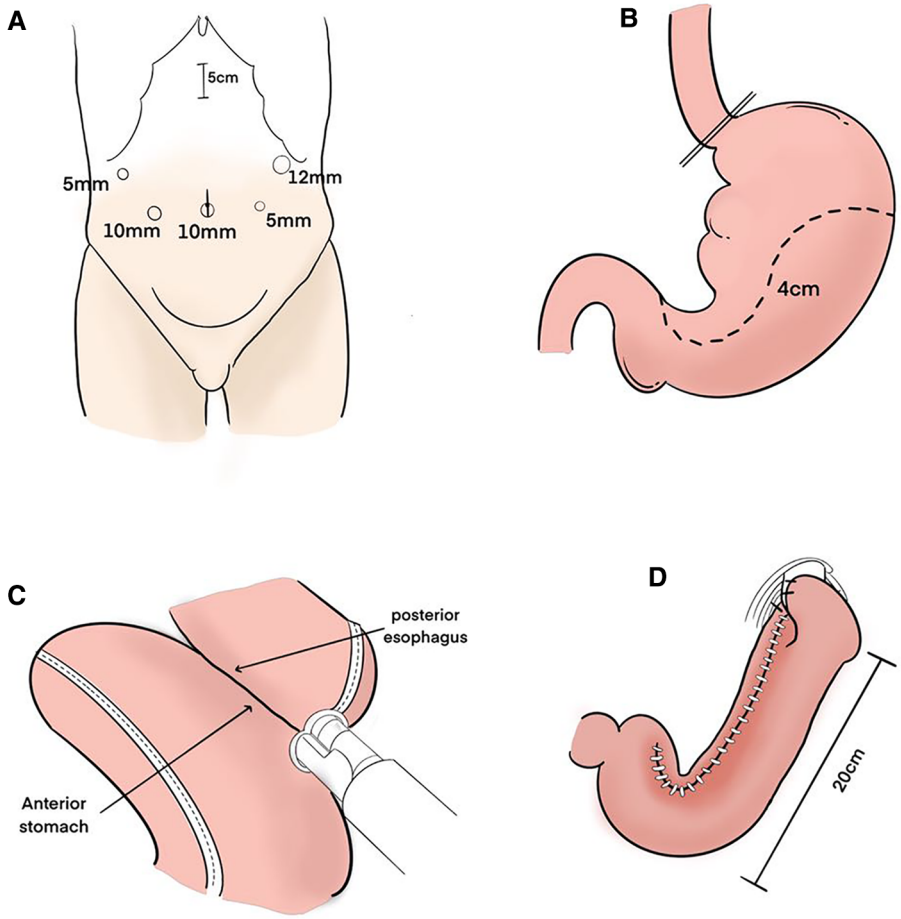


FIGURE 1
Surgical procedure. (A) Trocar sites. (B) After the esophagus was transected, the upper part of the stomach was excised along a dashed line to create a gastric tube. Its width is about 4 cm. (C) The anterior wall of the gastric tube and the posterior wall of the esophagus were anastomosed with linear stapler. (D) After anastomosis was performed, the entry hole was closed with barbed suture. The gastric tube was anchored with the right and left crus of the diaphragm by one stitch each to prevent hiatus hernia. The length of the whole gastric tube on the greater curvature side is about 20 cm.

abdomen, and the stomach was extruded through it. The stomach was marked with a dotted line using a sterile marker and ruler. Approximately 5 cm below the tumor, the upper part of the stomach was excised along a dashed line to make a gastric tube (Figure 1B). The pneumoperitoneum was re-established. An entry hole was made on the posterior side of the esophagus stump and on the anterior wall of the gastric tube 40 mm distal from the proximal stump laparoscopically. Then, the linear stapler was applied between the anterior wall of the gastric tube and the posterior wall of the esophagus laparoscopically (Figure 1C). After anastomosis was performed, the entrance hole was enclosed with stitches. The gastric tube was anchored with the right and left crus of the diaphragm by one stitch each to prevent hiatus hernia (Figure 1D). The vagus nerve was not preserved.

The length of the gastric tube is generally approximately 20 cm and the width is approximately 4 cm, which indicates: (1) The size of the residual stomach was correlated with gastric emptying; the larger the stomach, the worse the emptying; (2) Residual gastric emptying is related to gravity, intragastric pressure and residual gastric compliance; (3) The smaller the gastric tube, the worse its compliance is and the less likely it is to have reflux. The gastric tube after anastomosis with the esophagus is shown in (Figure 2A).

Reconstruction technique of esophagojejunostomy, the jejunum was transected at a distance of 20 cm from the Treus ligament, and a tube stapler was placed at a distance of 50 cm from the distal end by purse-string suture. The jejunum was anastomosed laterally through the proximal jejunum, and the stump was closed by GIA stapler, and the absorbable suture was embedded. The pneumoperitoneum was re-established,

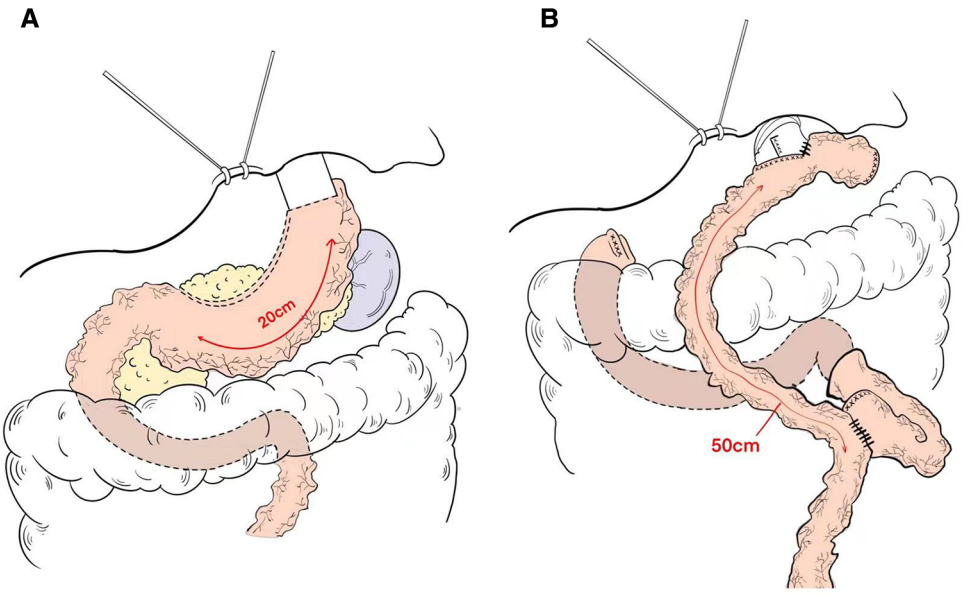


FIGURE 2
Schema of reconstruction after (A) proximal gastrectomy with tube gastric and (B) total gastrectomy with roux-en-Y esophagojejunostomy reconstruction.

and the esophagus and the distal jejunum were punctured. The lateral jejunum anastomosis of the posterior esophageal wall was completed by GIA stapler, and the common opening was closed by continuous suture of the barb line. Esophagojejunostomy (Roux-en-Y) was performed after total gastrectomy (**Figure 2B**).

## Data collection and evaluation of outcomes

Patients are enrolled in this study, three data management staff members will be assigned to collect relevant data. The basic characteristics of patients collected before surgery were age, sex, body mass index, ASA score, hematologic indices, serum tumor markers, FEV1 and Her2 expression. The clinicopathological features were TNM stage, tumor size and location, Lauren classification, and tumor cell differentiation. The operation was characterized by operative time, estimated blood loss, pathological proximal and distal margins, and number of lymph nodes removed. Postoperative outcomes were mean maximum body temperature during the first 3 days, analgesic use 1–5 days after surgery, days of bowel function recovery, time to start soft diet, postoperative hospital stay, and early complications (within 30 days after surgery). Patients were evaluated 1, 3, 6 and 12 months after operation, and their characteristics and results were obtained by viewing electronic medical records and picture archiving and communication systems. Postoperative morbidity was described based on the Clavien-Dindo classification of JOCG criteria for postoperative complications and according to the General Terminology criteria for Adverse Events (CTCAE 5.0) (21–23). Clinical features and nutritional status 1 year after operation were evaluated by PG-SGA (24).

Follow-up results 1 year postoperatively were based on the Visick classification (Grade i: asymptomatic; Grade ii: mild symptoms without medication; Grade iii: mild symptoms that are easily controlled by medication; Grade iiii: severe symptoms, the duration or surgery) assessment of reflux symptoms and endoscopic findings were scored in terms of the Los Angeles Classification of reflux esophagitis (25). The Visick score and endoscopy results were obtained in the clinic. All endoscopy results were graded by the same surgeon according to the Los Angeles Classification System.

Nutritional parameters after gastrectomy were assessed on the basis of changes in serum prealbumin, albumin, hemoglobin, prognostic nutritional index (PNI) and the percentage of BW loss (%BW loss) at 1, 3, 6, and 12 months after surgery (26). The percentage of BW loss (%BW loss) was calculated as follows: %BW loss = (BW at 1/3/6/12 months after surgery−preoperative BW)/(preoperative BW × 100). PNI was calculated using the following formula: 10 × serum albumin value (g/dl) + 0.005 × lymphocyte count in peripheral blood (27). On the CT images, the cross-sectional area of the psoas muscle was measured at the level of the third lumbar vertebra (L3). Psoas muscle index (PMI) = (Area of the psoas muscle at L3 [cm]2)/(height[m]2). %PMI loss was all defined in the same way as %BW loss (28, 29).

## Statistical analysis

Propensity score matching was based on gender, age, body mass index, clinical stage, and ASA score. Continuous variables of normal distribution were expressed by ($X \pm S$), and comparison between two groups was compared by $T$ test. Non-normally distributed continuous variables were represented by median (range), and comparisons between the two pairs were performed using Wilcoxon signed rank test. The categorical variables were expressed in terms of number of cases and percentage, and comparisons between the two groups were performed by Chi-square test or Fisher precise test. All analyses were performed using SPSS 26.0. ($P < 0.05$) was considered statistically significant.

## Ethics statement

The data for this study were collected in the course of general clinical practice, so informed consent signed by each patient was obtained for any surgical and clinical procedure. This protocol is in line with the ethical guidelines of the World Medical Association Declaration of Helsinki adopted by the 18th World Medical Association Congress held in Helsinki, Finland in June 1964. Institutional Review board approval is not required. Since this study was retrospective, patients' consent was not required for inclusion in the study.

# Results

## Patient characteristics

A total of 1,631 gastric cancer patients underwent surgery (Figure 1). Of those, 1,178 were excluded because the tumor was in the middle or lower part of the stomach. Of the remaining 453 patients, patients with advanced cancer ($n = 51$), neoadjuvant chemotherapy ($n = 48$) and dual primary cancer ($n = 3$). 59 patients received proximal gastrectomy and 292 patients received total gastrectomy. After 1:2 matching between the PG group and TG group, there were 54 patients in the PG group and 108 patients in the TG group (Figure 3 and Table 1).

The basic clinical characteristics of patients in the two groups were shown in Table 1. In the whole cohort, the number of patients who underwent ESD before surgery in the PG group was significantly greater than that in the TG group ($P = 0.011$), and there was no significant difference in preoperative hematological nutritional indicators, tumor markers or complications. The matched baseline features are well balanced.

## Perioperative clinical outcomes

In the entire cohort, the two groups were on a liquid diet ($4.11 \pm 1.77$ in PG vs. $4.91 \pm 1.89$; $P = 0.032$) and showed bowel function recovery ($2.77 \pm 1.25$ in PG vs. $3.44 \pm 1.62$ in TG; $P = 0.024$). The blood loss and operation time in the PG group were lower than those in TG group, but there were no significant differences ($P > 0.05$). There was no statistically significant difference in medical cost or 30-day readmission between the two groups ($P > 0.05$). After propensity score matching, the start of a soft diet after the operation in the PG group was 0.7 days sooner than in the TG group ($4.06 \pm 1.81$ vs. $4.76 \pm 1.69$; $P = 0.047$), and the recovery time of bowel function in the PG group was 0.72 days shorter than in the TG group ($2.29 \pm 1.16$ vs. $3.01 \pm 1.22$; $P = 0.039$). In the matched cohort, there were no statistically significant differences in the length of postoperative hospital stay, the average maximum body temperature in the first three days after surgery, and the number of patients using analgesics 1–5 days after surgery ($P > 0.05$) (Table 2).

## Histopathologic characteristic

Tumor size ($2.9 \pm 2.6$ vs. $9.4 \pm 4.2$), proximal resection margin ($2.5 \pm 1.4$ vs. $4.7 \pm 3.8$), distal resection margin ($3.1 \pm 1.9$ vs. $12 \pm 6.1$) and number of dissected lymph nodes ($18.31 \pm 9.49$ vs. $31.46 \pm 15.61$) were measured in the two groups, and the number of positive lymph nodes ($2.1 \pm 1.7$ vs. $7.32 \pm 3.17$), pTNM stage and histological type were significantly different ($P < 0.001$). Notably, the TG group accounted for 82.2% of the histological type of poorly differentiated adenocarcinoma. There were no significant differences in tumor location or Lauren classification between the two groups ($P > 0.05$). After propensity matching, there was no significant difference in pTNM staging between the two groups ($P > 0.05$), and tumors in the PG group were significantly smaller than those in the TG group ($2.3 \pm 1.2$ vs. $3.6 \pm 2.1$; $P = 0.027$). The proximal margin was smaller ($2.1 \pm 1.7$ vs. $4.1 \pm 3.6$; $P = 0.013$), lymph nodes were removed ($19.73 \pm 10.03$ vs. $24.75 \pm 12.84$; $P = 0.023$) and fewer were positive ($1.9 \pm 1.6$ vs. $3.7 \pm 4.1$; $P = 0.041$), and all differences were statistically significant (all $P < 0.05$) (Table 3).

## Operative complications and adverse events

Across the cohort, the severity of complications was classified by Clavien-Dindo and CTCAE version 5.0 classification. The number of early complications (i.e., complications occurring in the first 30 days after surgery) in

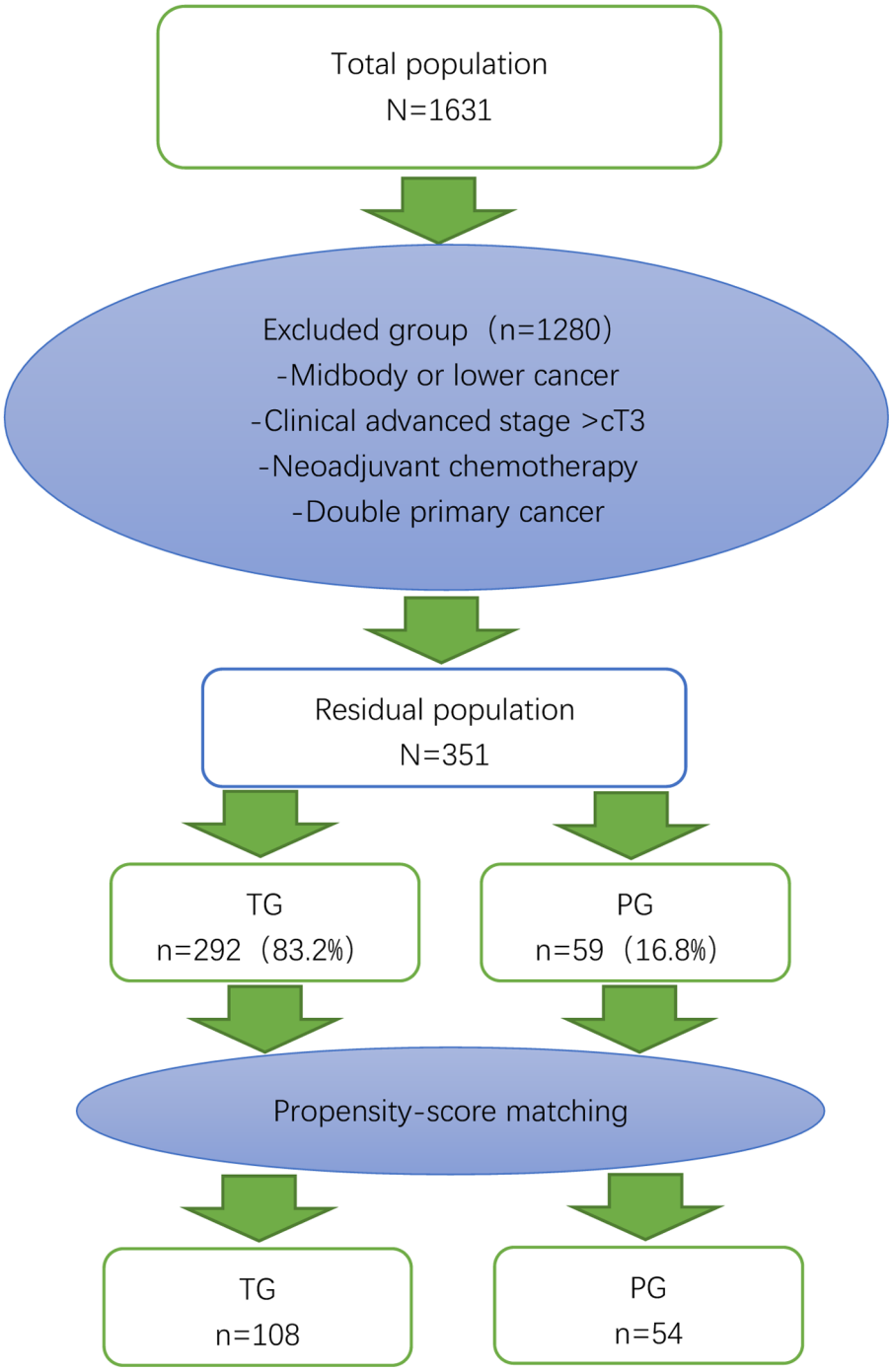


FIGURE 3
Flow chart of patient selection and propensity score matching. The 54 patients who underwent proximal gastrectomy (PG) were matched to 108 patients who underwent total gastrectomy (TG) in terms of age, sex, body mass index, clinical stage, and American Society of Anesthesiologists (ASA) score.

both groups (PG group 12 vs. TG group 64; $P = 0.788$) and hematological adverse events (36 in PG group vs. 188 in TG group; $P = 0.624$) were evaluated. Postoperative reflux occurred in 6 patients in the PG group and 13 patients in the TG group ($P = 0.076$), and severe complications (grade iii or above) occurred in 3 patients in the PG group and 13 patients in the TG group. Among the hematological serious adverse events, there were 2 cases in the PG group and 11 cases in

TABLE 1 Patient characteristics.

| Factor | Entire cohort | | *P* | Matched cohort | | *P* |
|---|---|---|---|---|---|---|
| | PG (*n* = 59) | TG (*n* = 292) | | PG (*n* = 54) | TG (*n* = 108) | |
| Age, year, median (range) | 63 (34–79) | 64 (29–78) | 0.524 | 65 (41–74) | 64 (39–73) | 0.879 |
| Sex | | | 0.096 | | | 0.303 |
| Male, *n* (%) | 42 (71.2) | 236 (80.8) | | 38 (70.4) | 84 (77.8) | |
| Female, *n* (%) | 17 (28.8) | 56 (19.2) | | 16 (29.6) | 24 (22.2) | |
| BMI, Kg/m$^2$ ± SD | 25.17 ± 3.24 | 24.35 ± 3.73 | 0.117 | 25.27 ± 3.08 | 24.91 ± 3.38 | 0.512 |
| ASA physical status | | | 0.322 | | | 0.480 |
| 0–1, *n* (%) | 21 (35.6) | 85 (29.1) | | 20 (37.0) | 34 (31.5) | |
| ≥2, *n* (%) | 38 (64.4) | 207 (70.9) | | 34 (63.0) | 74 (68.5) | |
| Preoperative Hb, g/L ± SD | 134.09 ± 14.93 | 131.01 ± 15.81 | 0.169 | 135.17 ± 15.14 | 132.46 ± 15.36 | 0.289 |
| Preoperative albumin, g/L ± SD | 43.01 ± 2.11 | 42.23 ± 2.43 | 0.022 | 42.57 ± 2.59 | 41.82 ± 2.85 | 0.106 |
| Psoas muscle index, cm$^2$/m$^2$ ± SD | 169.48 ± 33.09 | 168.49 ± 35.21 | 0.842 | 168.78 ± 32.13 | 167.52 ± 33.17 | 0.818 |
| Lymphocyte count, 10$^9$/L ± SD | 1.43 ± 0.39 | 1.46 ± 0.41 | 0.606 | 1.41 ± 0.45 | 1.44 ± 0.49 | 0.706 |
| Preoperative pre-albumin, g/L ± SD | 242.49 ± 36.02 | 234.41 ± 40.63 | 0.157 | 244.37 ± 35.13 | 237.09 ± 39.02 | 0.249 |
| Preoperative creatinine, umol/L ± SD | 61.03 ± 14.91 | 63.01 ± 15.17 | 0.360 | 62.98 ± 15.09 | 63.57 ± 13.31 | 0.800 |
| Previous treatment with ESD, *n* (%) | 8 (13.6) | 14 (4.8) | 0.011 | 8 (14.8) | 14 (12.9) | 0.746 |
| NRS 2002 Score | | | 0.406 | | | 0.652 |
| <3, *n* (%) | 36 (61.0) | 161 (55.1) | | 33 (61.1) | 62 (57.4) | |
| ≥3, *n* (%) | 23 (39.0) | 131 (44.9) | | 21 (38.9) | 46 (42.6) | |
| CCI score | | | 0.712 | | | 0.737 |
| 0–2, *n* (%) | 27 (45.8) | 126 (43.2) | | 25 (46.3) | 47 (43.5) | |
| ≥3, *n* (%) | 32 (54.2) | 166 (56.8) | | 29 (53.7) | 61 (56.5) | |
| Her2 | | | 0.391 | | | 0.554 |
| 0, *n* (%) | 43 (72.9) | 208 (71.2) | | 40 (74.1) | 78 (72.2) | |
| +, *n* (%) | 10 (16.9) | 58 (19.9) | | 9 (16.7) | 20 (18.5) | |
| ++, *n* (%) | 1 (1.7) | 14 (4.8) | | 1 (1.8) | 6 (5.6) | |
| +++, *n* (%) | 5 (8.5) | 12 (4.1) | | 4 (7.4) | 4 (3.7) | |
| AFP, ng/ml median (IQR) | 3.02 (2.28) | 2.88 (1.94) | 0.449 | 2.78 (2.21) | 2.91 (1.95) | 0.473 |
| CEA, ng/ml median (IQR) | 2.51 (2.74) | 2.76 (3.08) | 0.323 | 2.47 (2.63) | 2.67 (2.99) | 0.178 |
| CA-199, U/ml median (IQR) | 9.51 (9.68) | 10.56 (15.11) | 0.114 | 9.58 (9.81) | 11.12 (15.02) | 0.137 |
| CA-125, U/ml median (IQR) | 10.32 (6.40) | 9.78 (6.33) | 0.108 | 9.81 (6.63) | 9.94 (7.12) | 0.998 |
| CA-724, U/ml median (IQR) | 1.65 (2.36) | 2.32 (3.70) | 0.142 | 1.67 (2.42) | 2.09 (3.10) | 0.221 |
| CA-242, U/ml median (IQR) | 5.07 (4.09) | 5.44 (7.89) | 0.062 | 5.13 (4.70) | 5.59 (8.39) | 0.098 |
| History of smoking, *n* (%) | 31 (55.9) | 174 (59.6) | 0.317 | 29 (53.7) | 66 (61.1) | 0.367 |
| FEV1.0, % ± SD | 78.1 ± 10.3 | 77.3 ± 9.8 | 0.417 | 78.3 ± 10.6 | 77.6 ± 12 | 0.536 |
| Number of comorbidities | | | 0.640 | | | 0.884 |
| 0, *n* (%) | 24 (40.7) | 101 (34.6) | | 22 (40.7) | 39 (36.1) | |
| 1, *n* (%) | 27 (45.8) | 157 (53.8) | | 25 (46.3) | 56 (51.9) | |
| 2, *n* (%) | 6 (10.2) | 23 (7.9) | | 5 (9.3) | 10 (9.3) | |
| 3, *n* (%) | 2 (3.4) | 11 (3.8) | | 2 (3.7) | 3 (2.8) | |
| Comorbidities | | | 0.996 | | | 0.935 |
| Hypertension | 21 (35.6) | 109 (37.3) | | 19 (35.2) | 42 (38.9) | |
| Diabetes | 12 (20.3) | 66 (22.6) | | 10 (18.5) | 24 (22.2) | |
| Hepatic disease | 1 (1.7) | 5 (1.7) | | 1 (1.9) | 2 (1.9) | |
| Cardiac disease | 4 (6.8) | 19 (6.5) | | 2 (7.4) | 5 (4.6) | |
| Cerebrovascular disease | 4 (6.8) | 16 (5.5) | | 1 (5.6) | 4 (3.7) | |
| Asthma | 1 (1.7) | 5 (1.7) | | 1 (1.9) | 2 (1.9) | |
| History of pulmonary tuberculosis | 1 (1.7) | 4 (1.4) | | 0 | 1 (0.9) | |

TABLE 2 Perioperative indicators.

| Factor | Entire cohort | | *P* | Matched cohort | | *P* |
|---|---|---|---|---|---|---|
| | PG (*n* = 59) | TG (*n* = 292) | | PG (*n* = 54) | TG (*n* = 108) | |
| Operation time (min ± SD) | 166.41 ± 41.78 | 179.51 ± 42.84 | 0.032 | 165.32 ± 42.23 | 177.61 ± 42.96 | 0.086 |
| Estimated blood loss (ml ± SD) | 57.19 ± 27.81 | 64.86 ± 30.03 | 0.071 | 55.66 ± 4.49 | 57.21 ± 5.13 | 0.061 |
| Operation method | | | 0.120 | | | 0.081 |
| Laparoscopic, *n* (%) | 44 (74.6) | 187 (64.0) | | 40 (74.1) | 65 (60.2) | |
| Robotic, *n* (%) | 15 (25.4) | 105 (36.0) | | 14 (25.9) | 43 (39.8) | |
| Lymph node dissection, *n* (%) | | | <0.001 | | | <0.001 |
| D1+ | 58 (98.3) | 81 (27.7) | | 54 (100) | 62 (57.4) | |
| D2 | 1 (1.7) | 211 (72.3) | | 0 | 46 (42.6) | |
| Combined resection | | | 0.786 | | | >0.999 |
| Gallbladder | 2 | 6 | | 1 | 2 | |
| Spleen | 0 | 3 | | 0 | 1 | |
| Bowel function recovery (days ± SD) | 2.77 ± 1.25 | 3.19 ± 1.31 | 0.024 | 2.29 ± 1.16 | 2.71 ± 1.22 | 0.037 |
| Start of soft diet (days ± SD) | 4.11 ± 1.77 | 4.68 ± 1.89 | 0.033 | 4.06 ± 1.81 | 4.66 ± 1.69 | 0.039 |
| Analgesic use on Post-operative day 1-5, *n* (%) | 32 (54.2) | 167 (57.2) | 0.773 | 29 (53.7) | 61 (56.5) | 0.867 |
| Body temperature during the first 3 days[a] | | | | | | |
| Post-operative day 1 (mean ± SD) | 37.6°C ± 1.7°C | 37.8°C ± 1.9°C | 0.454 | 37.7°C ± 1.9°C | 37.9°C ± 1.7°C | 0.498 |
| Post-operative day 2 (mean ± SD) | 37.2°C ± 1.4°C | 37.3°C ± 1.3°C | 0.595 | 37.4°C ± 1.2°C | 37.3°C ± 1.5°C | 0.671 |
| Post-operative day 3 (mean ± SD) | 37.4°C ± 1.1°C | 37.3°C ± 1.6°C | 0.647 | 37.1°C ± 1.3°C | 37.2°C ± 1.4°C | 0.661 |
| Postoperative hospital stay (days ± SD) | 7.12 ± 6.39 | 7.51 ± 7.17 | 0.698 | 7.02 ± 6.86 | 7.71 ± 7.79 | 0.581 |
| 30-day reoperation, *n* (%) | 0 | 2 (0.68) | >0.999 | 0 | 0 | - |
| 30-day readmission, *n* (%) | 2 (3.4) | 11 (3.8) | >0.999 | 1 (1.9) | 4 (3.7) | 0.666 |
| Medical cost (*yuan* ± SD) | | | | | | |
| Laparoscopic | 70894.3 ± 2241 | 89912.6 ± 2873 | 0.132 | 71937.4 ± 2106 | 89644.6 ± 2923 | 0.276 |
| Robotic | 120773.7 ± 8796 | 131417.2 ± 5637 | 0.208 | 116030.2 ± 9022 | 120511.5 ± 4927 | 0.419 |

[a]The highest body temperature.

the TG group (grades 3–4). After matching, the number of complications in the two groups was 9 cases and 21 cases respectively ($P = 0.668$). Among the number of reflux cases, there were 6 cases in the PG group and 4 cases in the TG group ($P = 0.133$), showing no statistical significance. The number of adverse events was 32 cases and 71 cases ($P = 0.419$), among which, the number of anemia cases was 10 cases and 26 cases ($P = 0.423$), respectively, showing no statistically significant differences (Table 4).

## Clinical manifestations and nutritional status

There was no significant difference in clinical characteristics between the two groups 1 year after the operation. Overall, 26 patients in the PG group and 43 patients in the TG group ($P = 0.312$) reported no dietary problems. Reflux was present in 6 patients in the PG group and 3 patients in the TG group ($P = 0.069$), and there was no significant difference. In terms of nutrition score, although there was no statistical significance in different degrees of malnutrition between the two groups ($P = 0.406$), the PG group included a large proportion of mild malnutrition patients: 21 (38.9%) in the PG group and 33 (30.6%) in the TG group. Similarly, in the severe malnutrition patients, 2 (3.7%) were in the PG group. There were 9 patients in the TG group (8.3%) and a relatively small number in the PG group (Table 5).

We assessed the rate of weight loss, the rate of Psoas muscle index loss, and changes in nutritional parameters in 162 patients followed for at least 1 year (Figure 4). The annual decrease in serum hemoglobin in the TG group was greater than that in the PG group, and the decrease in serum hemoglobin in the TG group at 3 and 6 months after surgery was significantly higher than that in the PG group ($P = 0.032$ and 0.046, respectively), There was no significant difference between the two groups at 1 and 12 months after surgery ($P = 0.131$ and $P = 0.072$, respectively). Regarding PNI, there was no significant difference between the PG group and the TG group ($P > 0.05$). However, the PNI decline in the TG group was always higher than that in the PG group after surgery. Although the TG group recovered faster from 1 to 3 months,

TABLE 3 Histopathologic characteristics.

| Variable | Entire cohort | | *P* | Matched cohort | | *P* |
|---|---|---|---|---|---|---|
| | PG (*n* = 59) | TG (*n* = 292) | | PG (*n* = 54) | TG (*n* = 108) | |
| Tumor location, *n* (%) | | | 0.578 | | | 0.870 |
| EG junction | 15 (25.4) | 57 (19.5) | | 14 (25.9) | 22 (20.4) | |
| Cardia | 5 (8.5) | 27 (9.2) | | 5 (9.3) | 9 (8.3) | |
| Fundus | 21 (35.6) | 94 (32.2) | | 18 (33.3) | 41 (38.0) | |
| Upper body | 18 (30.5) | 114 (39.0) | | 17 (31.5) | 36 (33.3) | |
| Tumour size (cm ± SD) | 2.9 ± 2.6 | 9.4 ± 4.2 | <0.001 | 2.3 ± 1.2 | 3.6 ± 2.1 | 0.027 |
| Pathological proximal margin (cm ± SD) | 2.5 ± 1.4 | 4.7 ± 3.8 | <0.001 | 2.1 ± 1.7 | 4.1 ± 3.6 | 0.013 |
| Pathological distal margin (cm ± SD) | 3.1 ± 1.9 | 12 ± 6.1 | <0.001 | 2.4 ± 1.5 | 11 ± 5.2 | <0.001 |
| Histological type, *n* (%) | | | <0.001 | | | 0.166 |
| Poorly differentiated | 36 (61.0) | 240 (82.2) | | 32 (59.3) | 77 (71.3) | |
| Moderately differentiated | 18 (30.5) | 43 (14.7) | | 18 (33.3) | 28 (25.9) | |
| Well differentiated | 4 (6.8) | 2 (0.7) | | 3 (5.6) | 1 (0.9) | |
| Undifferentiated | 1 (1.7) | 7 (2.4) | | 1 (1.9) | 2 (1.9) | |
| Histology (Lauren classification), *n* (%) | | | 0.907 | | | 0.892 |
| Intestinal | 17 (28.8) | 82 (28.1) | | 15 (27.8) | 29 (26.9) | |
| Diffuse | 21 (35.6) | 93 (31.8) | | 20 (37.0) | 36 (33.3) | |
| Mixed | 18 (30.5) | 95 (32.5) | | 16 (29.6) | 38 (35.2) | |
| Indeterminate | 3 (5.1) | 22 (7.5) | | 3 (5.6) | 5 (4.6) | |
| T stage, *n* (%) | | | <0.001 | | | 0.989 |
| T1a | 23 (39.0) | 62 (21.2) | | 22 (40.7) | 42 (38.9) | |
| T1b | 21 (35.6) | 58 (19.9) | | 19 (35.2) | 38 (35.2) | |
| T2 | 14 (23.7) | 64 (21.9) | | 12 (22.2) | 26 (24.1) | |
| T3 | 1 (1.7) | 108 (37.0) | | 1 (1.9) | 2 (1.9) | |
| N stage, *n* (%) | | | <0.001 | | | 0.215 |
| N0 | 41 (69.5) | 139 (47.6) | | 39 (72.2) | 75 (69.4) | |
| N1 | 18 (30.5) | 78 (26.7) | | 15 (27.8) | 33 (30.6) | |
| N2 | 0 | 75 (25.7) | | 0 | 0 | |
| pTNM stage, *n* (%) | | | <0.001 | | | >0.999 |
| IA | 33 (55.9) | 86 (29.5) | | 32 (59.3) | 63 (58.3) | |
| IB | 22 (37.3) | 61 (20.9) | | 21 (38.9) | 43 (39.8) | |
| IIA | 4 (6.8) | 41 (14.0) | | 1 (1.9) | 2 (1.9) | |
| ≥IIB | 0 | 104 (35.6) | | 0 | 0 | |
| Retrieved lymph nodes (mean ± SD) | 18.31 ± 9.49 | 31.46 ± 15.61 | <0.001 | 19.73 ± 10.03 | 24.75 ± 12.84 | 0.023 |
| Positive lymph nodes (mean ± SD) | 2.1 ± 1.7 | 7.32 ± 3.17 | <0.001 | 1.9 ± 1.6 | 3.7 ± 4.1 | 0.041 |

the curves between the two groups have no intersection. Serum prealbumin levels in both groups were not significantly different at any time point ($P > 0.05$), but the TG group recovered faster at 3 to 6 months after surgery, and the levels in the two groups were almost the same at 12 months. Like albumin, there was no statistical significance at any time point, and the trend of change was not exactly the same as that of prealbumin. One month after surgery, %BW loss in TG group was significantly lower than that in the PG group ($P = 0.024$), and 6 months after surgery, %BW loss in the TG group was higher than that in the PG group. No significant differences were observed at any time point in % PMI loss between the two groups. No patients in either group died or relapsed during one year of follow-up.

## Reflux symptom and endoscopic findings

The patients will be graded according to the Visick score 1 year after operation, Visick score of reflux symptoms showed that there were 6 patients (11.1%) in the PG group and 3 patients (2.8%) in the TG group with grade ii or higher reflux symptoms ($P = 0.069$), which was not statistically significant. All patients underwent endoscopy approximately 1 year after

TABLE 4 Postoperative morbidity and adverse events within 30 postoperative days.

| Clavien-Dindo/CTCAE v5.0 | Entire cohort | | *P* | Matched cohort | | *P* |
|---|---|---|---|---|---|---|
| | PG (*n* = 59) | TG (*n* = 292) | | PG (*n* = 54) | TG (*n* = 108) | |
| Complications, *n* (%) | | | 0.788 | | | 0.668 |
| No | 47 (79.7) | 228 (78.1) | | 45 (83.3) | 87 (80.6) | |
| Yes | 12 (20.3) | 64 (21.9) | | 9 (16.7) | 21 (19.4) | |
| Clavien-Dindo Grade, *n* (%) | | | 0.928 | | | 0.939 |
| Grade I–II | 9 (15.3) | 51 (17.5) | | 8 (14.8) | 17 (15.7) | |
| Grade III–IV | 3 (5.1) | 13 (4.5) | | 1 (1.9) | 4 (3.7) | |
| Non-hematological, *n* (%) | | | 0.948 | | | 0.856 |
| Anastomotic leakage | 0 | 2 (0.7) | | 0 | 0 | |
| Anastomotic stenosis | 0 | 2 (0.7) | | 0 | 1 (0.9) | |
| Cholecystitis | 0 | 3 (1.0) | | 0 | 0 | |
| Pancreatitis | 1 (1.7) | 4 (1.4) | | 0 | 1 (0.9) | |
| Pancreatic fistula | 1 (1.7) | 5 (1.7) | | 1 (1.9) | 2 (1.9) | |
| Intraperitoneal hemorrhage | 1 (1.7) | 5 (1.7) | | 1 (1.9) | 2 (1.9) | |
| Fluid abscess | 0 | 7 (2.4) | | 0 | 1 (0.9) | |
| Wound infection | 0 | 4 (1.4) | | 0 | 1 (0.9) | |
| Wound dehiscence | 0 | 3 (1.0) | | 0 | 1 (0.9) | |
| Pneumonia | 2 (3.4) | 9 (3.1) | | 1 (1.9) | 5 (4.6) | |
| Chyle leakage | 1 (1.7) | 5 (1.7) | | 0 | 2 (1.9) | |
| Regurgitation | 6 (10.2) | 13 (4.5) | 0.077 | 6 (11.1) | 4 (3.7) | 0.133 |
| Ileus | 0 | 2 (0.7) | | 0 | 1 (0.9) | |
| Adverse events, *n* (%) | | | 0.624 | | | 0.419 |
| No | 23 (39.0) | 104 (35.6) | | 22 (40.7) | 37 (34.3) | |
| Yes | 36 (61.0) | 188 (64.4) | | 32 (59.3) | 71 (65.7) | |
| CTCAE v5.0 Grade, *n* (%) | | | 0.898 | | | 0.658 |
| Grade 1–2 | 34 (57.6) | 177 (60.6) | | 31 (57.4) | 67 (62.0) | |
| Grade 3–4 | 2 (3.4) | 11 (3.8) | | 1 (1.9) | 4 (3.7) | |
| Hematological, *n* (%) | | | 0.997 | | | 0.950 |
| Anemia[a] | 13 (22.0) | 74 (25.3) | 0.591 | 10 (16.7) | 26 (24.1) | 0.423 |
| Lymphocytopenia[b] | 1 (1.9) | 4 (1.4) | | 1 (1.9) | 1 (0.9) | |
| Creatinine increased[c] | 0 | 3 (1.0) | | 0 | 2 (1.9) | |
| Hypo-pre-albuminemia[d] | 12 (20.3) | 52 (17.8) | | 12 (22.2) | 27 (25.0) | |
| Hyperbilirubinemia[e] | 3 (5.1) | 21 (7.2) | | 3 (5.6) | 5 (4.6) | |
| AST/ALT increased[f] | 3 (5.1) | 14 (4.8) | | 2 (3.7) | 2 (1.9) | |
| Hypernatremia[g] | 0 | 1 (0.3) | | 0 | 0 | |
| Hyponatremia[h] | 3 (5.1) | 14 (4.8) | | 3 (7.4) | 7 (6.5) | |
| Hyperkalemia[i] | 1 (1.9) | 5 (1.7) | | 1 (1.9) | 1 (0.9) | |

[a]Male patients Hb < 110 g/L, female patients Hb < 100 g/L.
[b]Lymphocyte count < $1.1*10^9$/L.
[c]Creatinine > 132umol/L.
[d]Pre-albumin < 200 mg/L.
[e]Total bilirubin > 22umol/L.
[f]AST/ALT > 2.
[g]Na > 147 mmol/L.
[h]Na < 137 mmol/L.
[i]K > 5.3 mmol/L.

surgery. These endoscopic findings were scored against the Los Angeles classification of reflux esophagitis and the results of preoperative endoscopy. In the PG group, 5 patients had grade A reflux esophagitis and 1 patient had grade B reflux esophagitis before surgery. After surgery, 7 patients developed grade A reflux esophagitis and 1 patient developed grade B

TABLE 5 Comparison of postoperative clinical manifestations and nutritional score by PG-SGA between PG and TG 1 year after surgery.

| | PG ($n = 54$) | TG ($n = 108$) | *P* |
|---|---|---|---|
| Symptom, *n* (%) | | | 0.927 |
| There are no dietary problems | 26 (48.1) | 43 (39.8) | 0.312 |
| Nausea | 3 (5.6) | 7 (6.5) | |
| Mouth pain | 0 | 1 (0.9) | |
| A strange smell is scratching me | 1 (1.9) | 3 (2.8) | |
| Vomit | 2 (3.7) | 5 (4.6) | |
| Dry mouth | 2 (3.7) | 5 (4.6) | |
| No appetite | 2 (3.7) | 6 (5.6) | |
| Constipation | 2 (7.4) | 8 (7.4) | |
| Dysphagia | 1 (1.9) | 3 (2.8) | |
| Diarrhea | 1 (1.9) | 3 (2.8) | |
| Easy to fill | 5 (9.3) | 13 (12.0) | |
| It tastes tasteless or strange | 1 (1.9) | 3 (2.8) | |
| Abdominal pain | 2 (3.7) | 5 (4.6) | |
| Esophageal reflux | 6 (11.1) | 3 (1) | 0.069 |
| Overall evaluation, *n* (%) | | | 0.406 |
| Good nutritional status SGA-A (0–3) | 21 (38.9) | 33 (30.6) | 0.289 |
| Moderate or suspected malnutrition SGA-B (48) | 31 (57.4) | 66 (66.1) | 0.650 |
| Severe malnutrition SGA-C (>8) | 2 (3.7) | 9 (8.3) | 0.440 |

reflux. In the TG group, grade A, B, and C reflux esophagitis occurred preoperatively in 1, 5, and 2 patients, respectively. After surgery, 4, 1, and 2 patients had grade A, B, and C reflux esophagitis, respectively (**Tables 6**, **7**). After surgery, 8 patients (14.8%) in the PG group and 7 patients (6.5%) in the TG group had ≥A reflux ($P = 0.085$), showing no statistical significance.

## Discussion

Proximal gastrectomy was introduced to improve patient performance status by conserving half of the stomach; thus, it is widely believed that proximal gastrectomy reduces postoperative weight loss. In addition, PG in the upper third of the stomach was believed to be appropriate in terms of both its radicality and safety (30, 31).

Our study showed that PG gastric tubular reconstruction had the advantage of less postoperative anemia and less %BW loss than TG. This result is consistent with previous reports (9, 32). Some studies have reported that PG with double-tract reconstruction does not have any advantages for postoperative anemia (28, 33, 34). Our data showed no significant difference in total protein and serum prealbumin. This result was consistent with previous reports (9, 35, 36). Based on its safety and simplicity, we believe that gastric tubular reconstruction can be a viable option after proximal PG. The use of a gastric tube provides a simple and safe anastomosis for PG because it is a single anastomosis. Kitano et al. introduced a reconstruction method using a gastric tube after PG (37). The authors indicated that the technique was simple and less invasive for EGC in the upper third of the stomach and evaluate the effectiveness of gastric tubular reconstruction to prevent reflux after open PG (38).

In 2017, Toyomasu reported that gastric tubular reconstruction has advantages, including being less invasive compared to jejunal interposition, shorter surgical duration, less surgical blood loss, and maintenance of postoperative nutritional status (39). Some previous reports showed different types of complications of PG. RE is common complication after PG. In the present study, RE with symptoms was diagnosed in 6 (11.1%) of 54 patients. This result was almost compatible with previous reports (40–43). But the rate of RE (≥Los Angeles grade A) has been reported to be over 30% (36, 44). Chen et al. (40) reported that only 14.3% of patients showed reflux symptoms after tube gastric anastomosis, and 57% of patients exhibited reflux esophagitis. Compared with traditional esophagogastric anastomosis, this method has obvious antireflux advantages. Aihara et al. (45) showed that 14% of patients had reflux symptoms after tube gastric anastomosis, while the incidence of anastomotic stenosis was 35%. Ronellenfitsch et al. (46) demonstrated that the incidence of reflux symptoms was 21.4% early (1–6 months) after esophagogastrostomy and 33.3% long (>6 months) after esophagogastrostomy. However, the symptoms were mild. Endoscopic examination results revealed that 29% of patients had esophagitis, and only 2 of them had reflux symptoms. Another study reported that after 3 weeks to 1 year follow-up, gastric tube anastomosis in patients with reflux symptoms was lighter than traditional residual stomach esophagus anastomosis; however, after 2–10 years of follow-up, there was no statistically significant difference the rate of reflux symptoms in patients with in gastric tube esophagus anastomosis compared with traditional residual stomach esophagus anastomosis (47).

The reflux symptoms of all patients in this study were graded 1 year after surgery using the Visick score. In total, nine patients had grade II reflux symptoms. Notably, the PG patient with Visick grade II reflux did not exhibit signs of reflux esophagitis on endoscopy 1 year after surgery: the Los Angeles scores were both grade 0. In contrast, the fifteen patients who exhibited reflux esophagitis on endoscopy 1 year after surgery (their grades ranged from A to C) all had Visick grade I scores. Thus, reflux symptoms did not correlate well with endoscopic findings. Several other studies have also reported this, both in patients who underwent gastrectomy and in patients with gastroesophageal reflux disease (46, 48). This may reflect differences between individuals in terms of sensitivity to subjective symptoms. Further prospective studies

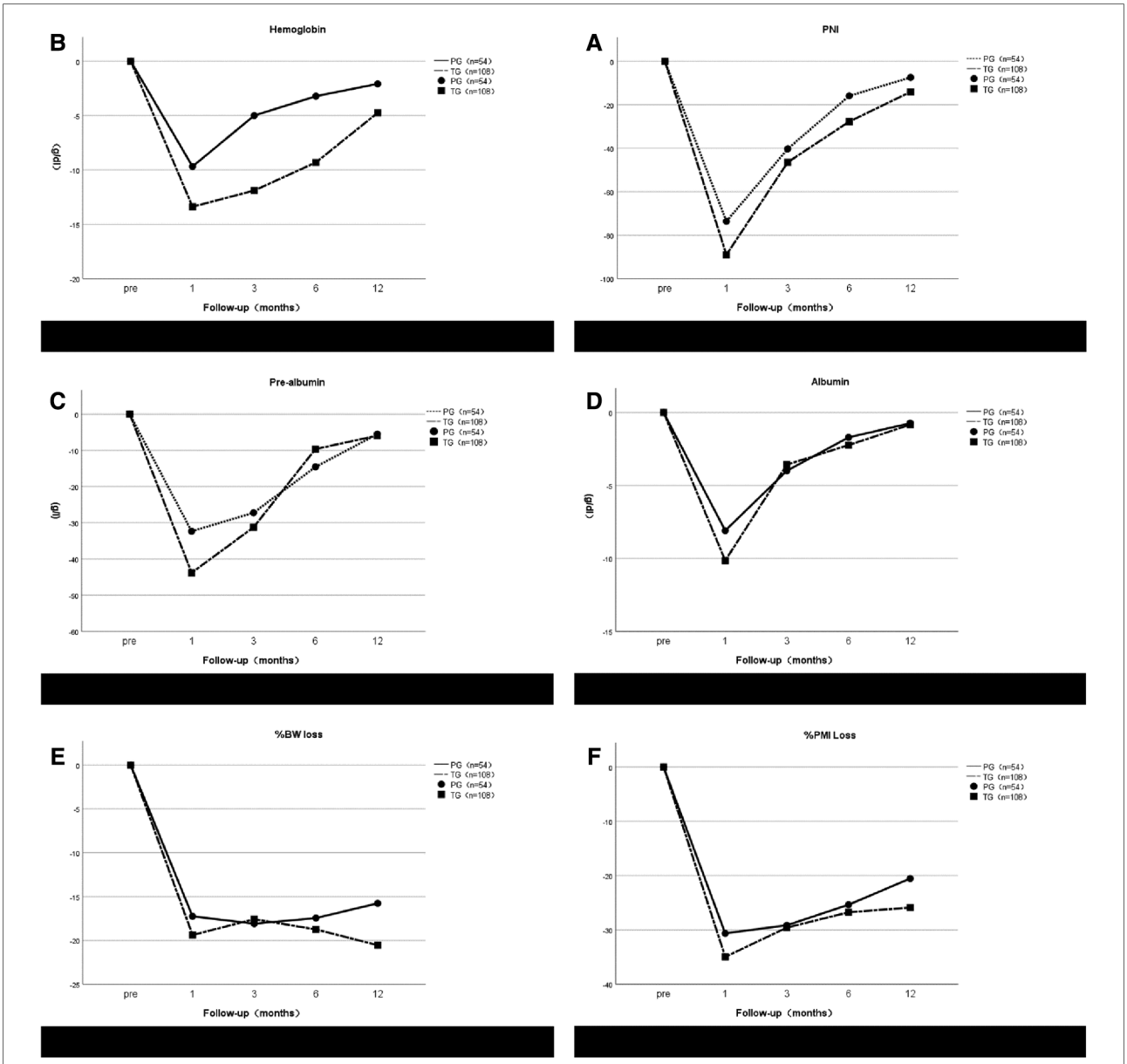


FIGURE 4
Postoperative changes of prognostic nutritional index (PNI) (**A**), hemoglobin (**B**), pre-albumin (**C**), albumin (**D**), %BW loss (**E**) and %PMI loss (**F**) in the proximal gastrectomy (PG) group and total gastrectomy (TG) group. All postoperative data are represented as values (mean ± standard error) relative to preoperative.

**TABLE 6** Reflux symptom scores 1 year after surgery in the propensity score-matched patients who underwent proximal or total gastrectomy.

| | PG ($n = 54$) | TG ($n = 108$) | $P$ |
|---|---|---|---|
| Visick score | | | 0.079 |
| I | 48 | 105 | 0.069 |
| II | 5 | 2 | 0.076 |
| III | 1 | 1 | >0.999 |
| IV | 0 | 0 | >0.999 |

on the relationship between reflux symptoms and reflux esophagitis on endoscopy are needed.

Recently, several useful assessment scales and questionnaires have been developed to measure the subjective reflux symptoms of patients. They include Post gastrectomy Syndrome Assessment Scale (PGSAS-45), Functional Assessment of Cancer Therapy-Gastric (FACT-Ga), and European Organization for Research and Treatment of Cancer Quality of Life Questionnaire-Gastric Cancer (EORTC QLQ-

TABLE 7 Endoscopic findings at 1 year after surgery.

| Reflux esophagitis | PG ($n = 54$) | | TG ($n = 108$) | |
|---|---|---|---|---|
| LA grade | Preoperative | Postoperative | Preoperative | Postoperative |
| A | 5 | 7 | 1 | 4 |
| B | 1 | 1 | 5 | 1 |
| C | 0 | 0 | 2 | 2 |
| D | 0 | 0 | 0 | 0 |

STO22 and EORTC QLQ-C30). Future studies comparing surgical modalities for EGC should use these tools to assess the postoperative functional benefits of each modality (8, 49, 50).

Multiple studies have described the advantages of PG for treating this cancer. However, to date, few studies have assessed the usefulness of PG with gastric tube placement. In particular, the oncological safety of PG with gastric tube placement remains unclear due to the lack of long-term studies. For PG with gastric tube to become the standard surgical option for early proximal gastric cancer, it must be as oncologically safe as TG, offer a functional benefit, and be associated with minimal postoperative complications.

The oncological safety of proximal gastrectomy mainly involves the preservation of the supratropyloric, supratropyloric, distal lesser curvature of the stomach, and lymph nodes along the right perivascular gastroomentum. Studies have shown that there is no statistically significant difference in the overall postoperative survival rate between patients undergoing total gastrectomy and patients undergoing proximal gastrectomy for early upper gastric cancer (51). Therefore, the clinical oncology safety of proximal gastrectomy for early upper gastric cancer is not controversial, but for advanced upper gastric cancer, the oncology safety of proximal gastrectomy is still controversial. Oncology safety depends primarily on the impact of lymph node preservation on patient survival.

Yamashita et al. (52) showed that for esophageal and gastric junction carcinoma with tumor length <4 cm, the lymph node metastasis rates of Groups 4sa, 4sb, 4d, 5 and 6 were extremely low and were independent of tumor location and T stage. The results of a separate study of 202 patients with stage T2 and T3 proximal gastric cancer undergoing proximal gastrectomy demonstrated that the lymph node metastasis rates of Groups 4sa, 4sb, 4d, 5, 6, 8a and 12a were 3.47%, 1.49%, 0.99%, 0.00%, 0.00%, 2.02% and 0.006%, respectively, and the overall 5-year survival rate of the patients was 72.9%. Proximal gastrectomy was recommended for the treatment of T2 and T3 proximal gastric cancer (53). However, no prospective randomized controlled studies have been conducted on the long-term outcomes of total gastrectomy and proximal gastrectomy for locally advanced upper gastric cancer, and the oncological safety of proximal gastrectomy needs further clinical evidence.

There are several limitations to this study. First, the small number of patients in the cohort and the retrospective design of this study made the evidence of retrospective analysis less reliable than that of randomized controlled trials. These limitations were offset using propensity score matching to select a TG group that matched the PG group in terms of important baseline features. Even though propensity score matching was used, there is a selection bias according to the preference of the surgeon. However, surgery performed during the same period and the proficiency of the surgical method are not different. Second, the lack of long-term follow-up of nutritional status, assessment of patients' long-term quality of life, and oncology outcomes largely limit the true benefits of PG. Third, we did not compare surgical methods in terms of postoperative VitB12 and serum iron, and anemia is a common complication of gastrectomy (54). Future studies comparing PG with tube gastric versus TG should examine postoperative iron panel blood test results. These studies should also determine the nutritional benefit of the surgical modalities by assessing the postoperative lipid profile. Finally, different reflux-scoring tools are needed to determine the effect of the surgeries on reflux symptoms.

## Conclusion

In this study, proximal gastrectomy with tubular gastrostomy was superior in clinical outcome to Roux-en-Y reconstruction for gastric cancer. These results suggest that PG combined with gastric tube anastomosis may be an appropriate surgical option for proximal gastric cancer. However, there is no standard procedure for early upper gastric cancer, and only prospective randomized trials in the future will clarify the true benefits of one procedure over another.

## Data availability statement

The original contributions presented in the study are included in the article/Supplementary Material, further inquiries can be directed to the corresponding author/s.

## Ethics statement

Written informed consent was obtained from the individual(s) for the publication of any potentially identifiable images or data included in this article.

## Author contributions

ZN made substantial contributions to conception and design for this study, YL and XJ acquired and analyzed data and JF drafted the article. HQ and YW gave many important suggestions for this study. ZN and XL participated in interpreting critically for important intellectual results. ZN gave final approval of the version to be published. All authors contributed to the article and approved the submitted version.

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## Conflict of interest

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

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## References

1. Kusano C, Gotoda T, Khor CJ, Katai H, Kato H, Taniguchi H, et al. Changing trends in the proportion of adenocarcinoma of the esophagogastric junction in a large tertiary referral center in Japan. *J Gastroenterol Hepatol.* (2008) 23 (11):1662–5. doi: 10.1111/j.1440-1746.2008.05572.x

2. Buas MF, Vaughan TL. Epidemiology and risk factors for gastroesophageal junction tumors: understanding the rising incidence of this disease. *Semin Radiat Oncol.* (2013) 23(1):3–9. doi: 10.1016/j.semradonc.2012.09.008

3. Information Committee of Korean Gastric Cancer Association. Korean gastric cancer association nationwide survey on gastric cancer in 2014. *J Gastric Cancer.* (2016) 16(3):131–40. doi: 10.5230/jgc.2016.16.3.131

4. Liu K, Yang K, Zhang W, Chen X, Chen X, Zhang B, et al. Changes of esophagogastric junctional adenocarcinoma and gastroesophageal reflux disease among surgical patients during 1988-2012: a single-institution, high-volume experience in China. *Ann Surg.* (2016) 263(1):88–95. doi: 10.1097/SLA.0000000000001148

5. Japanese Gastric Cancer Association. Japanese gastric cancer treatment guidelines 2018 (5th edition). *Gastric Cancer.* (2021) 24(1):1–21. doi: 10.1007/s10120-020-01042-y

6. Fujiya K, Kawamura T, Omae K, Makuuchi R, Irino T, Tokunaga M, et al. Impact of malnutrition after gastrectomy for gastric cancer on long-term survival. *Ann Surg Oncol.* (2018) 25(4):974–83. doi: 10.1245/s10434-018-6342-8

7. Rosa F, Quero G, Fiorillo C, Bissolati M, Cipollari C, Rausei S, et al. Total vs proximal gastrectomy for adenocarcinoma of the upper third of the stomach: a propensity-score-matched analysis of a multicenter western experience (on behalf of the Italian research group for gastric cancer-GIRCG). *Gastric Cancer.* (2018) 21(5):845–52. doi: 10.1007/s10120-018-0804-3

8. Takiguchi N, Takahashi N, Ikeda M, Inagawa S, Ueda S, Nobuoka T, et al. Long-term quality-of-life comparison of total gastrectomy and proximal gastrectomy by postgastrectomy syndrome assessment scale (PGSAS-45): a nationwide multi-institutional study. *Gastric Cancer.* (2015) 18(2):407–16. doi: 10.1007/s10120-014-0377-8

9. Ichikawa D, Komatsu S, Kubota T, Okamoto K, Shiozaki A, Fujiwara H, et al. Long-term outcomes of patients who underwent limited proximal gastrectomy. *Gastric Cancer.* (2014) 17(1):141–5. doi: 10.1007/s10120-013-0257-7

10. Shi M, Hu Z, Wu K, Yang D, Fu H, Zhang J, et al. Comparative study of pyloromyotomy and H-M pyloroplasty in proximal gastrectomy for adenocarcinoma of esophageal-gastric junction. *J Gastrointest Surg.* (2022) 26 (8):1585–95. doi: 10.1007/s11605-022-05347-4

11. Ichikawa D, Komatsu S, Okamoto K, Shiozaki A, Fujiwara H, Otsuji E, et al. Evaluation of symptoms related to reflux esophagitis in patients with esophagogastrostomy after proximal gastrectomy. *Langenbecks Arch Surg.* (2013) 398(5):697–701. doi: 10.1007/s00423-012-0921-0

12. Jung DH, Ahn SH, Park DJ, Kim HH. Proximal gastrectomy for gastric cancer. *J Gastric Cancer.* (2015) 15(2):77–86. doi: 10.5230/jgc.2015.15.2.77

13. Shoji Y, Nunobe S, Ida S, Kumagai K, Ohashi M, Sano T, et al. Surgical outcomes and risk assessment for anastomotic complications after laparoscopic proximal gastrectomy with double-flap technique for upper-third gastric cancer. *Gastric Cancer.* (2019) 22(5):1036–43. doi: 10.1007/s10120-019-00940-0

14. Sato R, Kinoshita T, Akimoto E, Yoshida M, Nishiguchi Y, Harada J, et al. Feasibility and quality of life assessment of laparoscopic proximal gastrectomy using double-tract reconstruction. *Langenbecks Arch Surg.* (2021) 406(2):479–89. doi: 10.1007/s00423-020-02076-7

15. Aburatani T, Kojima K, Otsuki S, Murase H, Okuno K, Gokita K, et al. Double-tract reconstruction after laparoscopic proximal gastrectomy using detachable ENDO-PSD. *Surg Endosc.* (2017) 31(11):4848–56. doi: 10.1007/s00464-017-5539-4

16. Kinoshita T, Gotohda N, Kato Y, Takahashi S, Konishi M, Kinoshita T, et al. Laparoscopic proximal gastrectomy with jejunal interposition for gastric cancer in the proximal third of the stomach: a retrospective comparison with open surgery. *Surg Endosc.* (2013) 27(1):146–53. doi: 10.1007/s00464-012-2401-6

17. Shiraishi N, Hirose R, Morimoto A, Kawano K, Adachi Y, Kitano S, et al. Gastric tube reconstruction prevented esophageal reflux after proximal gastrectomy. *Gastric Cancer.* (1998) 1(1):78–9. doi: 10.1007/s101200050058

18. Toyomasu Y, Mochiki E, Ishiguro T, Ito T, Suzuki O, Ogata K, et al. Clinical outcomes of gastric tube reconstruction following laparoscopic proximal gastrectomy for early gastric cancer in the upper third of the stomach: experience with 100 consecutive cases. *Langenbecks Arch Surg.* (2021) 406 (3):659–66. doi: 10.1007/s00423-021-02132-w

19. Sano T, Aiko T. New Japanese classifications and treatment guidelines for gastric cancer: revision concepts and major revised points. *Gastric Cancer.* (2011) 14(2):97–100. doi: 10.1007/s10120-011-0040-6

20. Brierley JD GM, Wittekind C. *TNM classification of malignant tumours*. 8th ed. New Jersey: Wiley Blackwell (2017).

21. Dindo D, Demartines N, Clavien PA. Classification of surgical complications: a new proposal with evaluation in a cohort of 6336 patients and results of a survey. *Ann Surg*. (2004) 240(2):205–13. doi: 10.1097/01.sla.0000133083.54934.ae

22. Katayama H, Kurokawa Y, Nakamura K, Ito H, Kanemitsu Y, Masuda N, et al. Extended clavien-dindo classification of surgical complications: japan clinical oncology group postoperative complications criteria. *Surg Today*. (2016) 46(6):668–85. doi: 10.1007/s00595-015-1236-x

23. Institute NC. Common terminology criteria for adverse events (CTCAE) version 5.0. Bethesda: National Cancer Institute (2017).

24. De Groot LM, Lee G, Ackerie A, van der Meij BS. Malnutrition screening and assessment in the cancer care ambulatory setting: mortality predictability and validity of the patient-generated subjective global assessment short form (PG-SGA SF) and the GLIM criteria. *Nutrients*. (2020) 12(8):1–18. doi: 10.3390/nu12082287

25. Kubo M, Sasako M, Gotoda T, Ono H, Fujishiro M, Saito D, et al. Endoscopic evaluation of the remnant stomach after gastrectomy: proposal for a new classification. *Gastric Cancer*. (2002) 5(2):83–9. doi: 10.1007/s101200200014

26. Demirelli B, Babacan N, Ercelep Ö, Öztürk M, Kaya S, Tanrıkulu E, et al. Modified Glasgow prognostic score, prognostic nutritional index and ECOG performance score predicts survival better than sarcopenia, cachexia and some inflammatory indices in metastatic gastric cancer. *Nutr Cancer*. (2021) 73 (2):230–8. doi: 10.1080/01635581.2020.1749290

27. Furukawa H, Kurokawa Y, Takiguchi S, Tanaka K, Miyazaki Y, Makino T, et al. Short-term outcomes and nutritional status after laparoscopic subtotal gastrectomy with a very small remnant stomach for cstage I proximal gastric carcinoma. *Gastric Cancer*. (2018) 21(3):500–7. doi: 10.1007/s10120-017-0755-0

28. Sugiyama M, Oki E, Ando K, Nakashima Y, Saeki H, Maehara Y, et al. Laparoscopic proximal gastrectomy maintains body weight and skeletal muscle better than total gastrectomy. *World J Surg*. (2018) 42(10):3270–6. doi: 10.1007/s00268-018-4625-7

29. Asaoka R, Irino T, Makuuchi R, Tanizawa Y, Bando E, Kawamura T, et al. Changes in body weight, skeletal muscle and adipose tissue after gastrectomy: a comparison between proximal gastrectomy and total gastrectomy. *ANZ J Surg*. (2019) 89(1-2):79–83. doi: 10.1111/ans.15023

30. Katai H, Sano T, Fukagawa T, Shinohara H, Sasako M. Prospective study of proximal gastrectomy for early gastric cancer in the upper third of the stomach. *Br J Surg*. (2003) 90(7):850–3. doi: 10.1002/bjs.4106

31. Shiraishi N, Adachi Y, Kitano S, Kakisako K, Inomata M, Yasuda K, et al. Clinical outcome of proximal versus total gastrectomy for proximal gastric cancer. *World J Surg*. (2002) 26(9):1150–4. doi: 10.1007/s00268-002-6369-6

32. Hayami M, Hiki N, Nunobe S, Mine S, Ohashi M, Kumagai K, et al. Clinical outcomes and evaluation of laparoscopic proximal gastrectomy with double-flap technique for early gastric cancer in the upper third of the stomach. *Ann Surg Oncol*. (2017) 24(6):1635–42. doi: 10.1245/s10434-017-5782-x

33. Cho M, Son T, Kim H, Noh S, Choi S, Seo W, et al. Similar hematologic and nutritional outcomes after proximal gastrectomy with double-tract reconstruction in comparison to total gastrectomy for early upper gastric cancer. *Surg Endosc*. (2019) 33(6):1757–68. doi: 10.1007/s00464-018-6448-x

34. Park JY, Park KB, Kwon OK, Yu w. Comparison of laparoscopic proximal gastrectomy with double-tract reconstruction and laparoscopic total gastrectomy in terms of nutritional status or quality of life in early gastric cancer patients. *Eur J Surg Oncol*. (2018) 44(12):1963–70. doi: 10.1016/j.ejso.2018.08.014

35. Ahn SH, Lee JH, Park DJ, Kim HH. Comparative study of clinical outcomes between laparoscopy-assisted proximal gastrectomy (LAPG) and laparoscopy-assisted total gastrectomy (LATG) for proximal gastric cancer. *Gastric Cancer*. (2013) 16(3):282–9. doi: 10.1007/s10120-012-0178-x

36. Chen S, Li J, Liu H, Zeng J, Yang G, Wang J, et al. Esophagogastrostomy plus gastrojejunostomy: a novel reconstruction procedure after curative resection for proximal gastric cancer. *J Gastrointest Surg*. (2014) 18(3):497–504. doi: 10.1007/s11605-013-2391-2

37. Kitano S, Adachi Y, Shiraishi N, Suematsu T, Bando T. Laparoscopic-assisted proximal gastrectomy for early gastric carcinomas. *Surg Today*. (1999) 29(4):389–91. doi: 10.1007/BF02483072

38. Adachi Y, Inoue T, Hagino Y, Shiraishi N, Shimoda K, Kitano S, et al. Surgical results of proximal gastrectomy for early-stage gastric cancer: jejunal interposition and gastric tube reconstruction. *Gastric Cancer*. (1999) 2(1):40–5. doi: 10.1007/s101200050019

39. Toyomasu Y, Ogata K, Suzuki M, Yanoma T, Kimura A, Kogure N, et al. Restoration of gastrointestinal motility ameliorates nutritional deficiencies and body weight loss of patients who undergo laparoscopy-assisted proximal gastrectomy. *Surg Endosc*. (2017) 31(3):1393–401. doi: 10.1007/s00464-016-5127-z

40. Chen XF, Zhang B, Chen ZX, Hu JK, Dai B, Wang F, et al. Gastric tube reconstruction reduces postoperative gastroesophageal reflux in adenocarcinoma of esophagogastric junction. *Dig Dis Sci*. (2012) 57(3):738–45. doi: 10.1007/s10620-011-1920-7

41. Hosogi H, Yoshimura F, Yamaura T, Satoh S, Uyama I, Kanaya S, et al. Esophagogastric tube reconstruction with stapled pseudo-fornix in laparoscopic proximal gastrectomy: a novel technique proposed for siewert type II tumors. *Langenbecks Arch Surg*. (2014) 399(4):517–23. doi: 10.1007/s00423-014-1163-0

42. Yasuda A, Yasuda T, Imamoto H, Kato H, Nishiki K, Iwama M, et al. A newly modified esophagogastrostomy with a reliable angle of his by placing a gastric tube in the lower mediastinum in laparoscopy-assisted proximal gastrectomy. *Gastric Cancer*. (2015) 18(4):850–8. doi: 10.1007/s10120-014-0431-6

43. Ueda Y, Shiraishi N, Toujigamori M, Shiroshita H, Etoh T, Inomata M, et al. Laparoscopic proximal gastrectomy with gastric tube reconstruction. *JSLS*. (2016) 20(3):1–8. doi: 10.4293/JSLS.2016.00046

44. Tokunaga M, Ohyama S, Hiki N, Hoshino E, Nunobe S, Fukunaga T, et al. Endoscopic evaluation of reflux esophagitis after proximal gastrectomy: comparison between esophagogastric anastomosis and jejunal interposition. *World J Surg*. (2008) 32(7):1473–7. doi: 10.1007/s00268-007-9459-7

45. Aihara R, Mochiki E, Ohno T, Yanai M, Toyomasu Y, Ogata K, et al. Laparoscopy-assisted proximal gastrectomy with gastric tube reconstruction for early gastric cancer. *Surg Endosc*. (2010) 24(9):2343–8. doi: 10.1007/s00464-010-0947-8

46. Ronellenfitsch U, Najmeh S, Andalib A, Perera R, Rousseau M, Mulder D, et al. Functional outcomes and quality of life after proximal gastrectomy with esophagogastrostomy using a narrow gastric conduit. *Ann Surg Oncol*. (2015) 22(3):772–9. doi: 10.1245/s10434-014-4078-7

47. Zhang M, Zhang C, Wu QC. Health-related quality of life and survival among 10-year survivors of esophageal cancer surgery: gastric tube reconstruction versus whole stomach reconstruction. *J Thorac Dis*. (2019) 11 (8):3284–91. doi: 10.21037/jtd.2019.08.56

48. Fennerty MB, Johnson DA. Heartburn severity does not predict disease severity in patients with erosive esophagitis. *MedGenMed*. (2006) 8(2):6. PMID:16926745; PMCID: PMC1785158

49. Garland SN, Pelletier G, Lawe A, Biagioni B, Easaw J, Eliasziw M, et al. Prospective evaluation of the reliability, validity, and minimally important difference of the functional assessment of cancer therapy-gastric (FACT-ga) quality-of-life instrument. *Cancer*. (2011) 117(6):1302–12. doi: 10.1002/cncr.25556

50. Blazeby JM, Conroy T, Bottomley A, Vickery C, Arraras J, Sezer O, et al. Clinical and psychometric validation of a questionnaire module, the EORTC QLQ-STO 22, to assess quality of life in patients with gastric cancer. *Eur J Cancer*. (2004) 40(15):2260–8. doi: 10.1016/j.ejca.2004.05.023

51. Xu Y, Tan Y, Wang YB, Xi C, Ye NY, Xu XZ, et al. Proximal versus total gastrectomy for proximal early gastric cancer: a systematic review and meta-analysis. *Medicine*. (2019) 98(19):e15663. doi: 10.1097/MD.0000000000015663

52. Yamashita H, Seto Y, Sano T, Makuuchi H, Ando N, Sasako M, et al. Results of a nation-wide retrospective study of lymphadenectomy for esophagogastric junction carcinoma. *Gastric Cancer*. (2017) 20(Suppl 1):69–83. doi: 10.1007/s10120-016-0663-8

53. Yura M, Yoshikawa T, Otsuki S, Yamagata Y, Morita S, Katai H, et al. Oncological safety of proximal gastrectomy for T2/T3 proximal gastric cancer. *Gastric Cancer*. (2019) 22(5):1029–35. doi: 10.1007/s10120-019-00938-8

54. Lee JH, Hyung WJ, Kim H, Kim YM, Son T, Okumura N, et al. Method of reconstruction governs iron metabolism after gastrectomy for patients with gastric cancer. *Ann Surg*. (2013) 258(6):964–9. doi: 10.1097/SLA.0b013e31827eebc1

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# High and low inferior mesenteric artery ligation in laparoscopic low anterior rectal resections: A retrospective study

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**Backgroud:** The high or low inferior mesenteric artery (IMA) ligation in rectal cancer remains a great debate. This study retrospectively discussed the outcomes of the perioperative period, defecation and urinary function and long-term prognosis in rectal cancer patients with high or low IMA ligation.
**Methods:** This study enrolled 220 consecutive rectal cancer cases, including 134 with high IMA ligation and 86 with low ligation. A comparison between the two groups was made for anastomotic leakage, low anterior resection syndrome (LARS), international prostate symptom score (IPSS), 5-year disease-free survival (DFS) and 5-year overall survival (OS).
**Results:** Low-ligation group had a longer operative time, and larger intraoperative blood loss. No significant difference was noted in anastomotic leakage incidence. In multivariable analysis, the male gender and tumor located at the lower rectum were identified as risk factors for anastomotic leakage. No significant differences were observed between groups in their LARS and IPSS questionnaire responses. The high-ligation vs. the low-ligation 5-year OS and DFS were 78.3% vs. 82.4% and 72.4% vs. 76.6%, respectively, which were not statistically different.
**Conclusion:** The ligation level of the IMA had no significant effect on the anastomotic leakage incidence, defecation, urinary function, and long-term prognosis.

KEYWORDS
rectal cancer, high ligation, low ligation, inferior mesenteric artery (IMA), anastomotic leakage (AL)

## Introduction

Currently, the incidence and mortality attributed to colorectal cancer both rank third, with patients becoming younger in average age (1). Surgical treatment is the mainstay for rectal cancer, amongst which low anterior resections are most valued. In domestic practice and overseas, there remains debate about the position of the inferior mesenteric artery (IMA) ligation during surgery (2–5). High ligation refers to a ligation located 1 to 2 cm away from the abdominal aorta origin, without the left colic artery preservation. In contrast, low ligation refers to a ligation at the left colic artery region, with dissection of the lymph nodes at IMA root and preservation of the left colic artery (6–9). Some scholars supported the use of high ligation mainly for two reasons: on the one hand,

high ligation allows a sufficient length of the proximal free colon, which ensures a situation free of tension for colonic anastomosis; on the other hand, high ligation raises the lymph node yield, and improves the precision of disease staging, in the way of allowing lymph nodes dissection at IMA root to the maximum (10–13). However, according to some scholars, high ligation is at the cost of abandoning proximal colonic blood perfusion and declining anastomotic blood supply, which might increase the incidence of anastomotic leakage and lead to colon necrosis due to ischemia in severe cases (14–17). Researchers have not yet discovered if high ligation contributes to a higher lymph node yield in rectal resections, while it shows no superiority to low ligation regarding long-term oncologic prognosis (8, 18, 19).

In addition, high ligation with complete lymph node IMA root dissection may damage the inferior epigastric plexus, which governs defecation, urination, and sexual function. The results are conflicting on the effect of IMA ligation level on defecation and urinary function (20, 21).

This study retrospectively discussed the outcomes of the perioperative period, defecation, urinary function, and long-term prognosis in rectal cancer patients who had a laparoscopic lower rectal anterior resection with high or low IMA ligation.

# Methods

## Patients

This retrospective study involved 220 participants treated with radical laparoscopic for rectal cancer in the Second Affiliated Hospital of Chongqing Medical University between January 2014 and May 2016. Inclusion criteria were (1) Distance between tumor edge and the anal verge ≤15 cm; (2) Preoperative colonoscopy confirmed rectal tumor, and the pathological tissue biopsy revealed adenocarcinoma; (3) Preoperative chest and abdomen computer tomography (CT) and pelvic MRI confirmed locally progressive tumor; (4) Intraoperative sigmoid-rectal end-to-end anastomosis with a double stapling apparatus. Exclusion criteria were (1) Stage IV carcinoma with distant or peritoneal metastases before or during operation; (2) Emergency patients complicated with bleeding, perforation, and intestinal obstruction; (3) Patients with Hartmann's or abdominoperineal resections; (4) Multiple colorectal cancers. The patient whose preoperative MRI suggested T3–4 or N+ was treated with concurrent radiotherapy and chemotherapy. The long course of preoperative radiotherapy consisted of 50.4 Gray in 25 to 28 fractions, five times per week, over five weeks. Oral 5-fluorouracil (Capecitabin, Xeloda®, 825 mg/m$^2$/day, twice a day, five times a week, for five weeks) was administered in conjunction with a long course of radiotherapy. After concurrent chemoradiotherapy, surgery was performed six weeks later. The hospital ethics committee approved this research.

## Surgical procedure

All surgeries were performed by five professors, each with 20 years of experience in gastrointestinal surgery, who decided on the ligation level of IMA according to the intraoperative situation and personal opinions. IMA was ligated 1 to 2 cm distant from the aorta origin, with clearance of lymph nodes at the root in the high-ligation cases. IMA was ligated in the lower part of the left colic artery following lymph node dissection at the root in the low-ligation cases. Following the sigmoid-rectal end-to-end anastomosis completion, one drainage tube was placed around the pelvic anastomosis and passed out through the abdominal wall. The surgeon decided the level of IMA ligation, whether to perform a preventive ileostomy, and whether to place an anal canal.

## Postoperative management

Training for bladder function was arranged by clamping the urinary catheter on day 2 after the operation, followed by removing the urinary catheter. The anal canal was removed on day 5. The abdominal drainage tube was removed on day 7 in case of the absence of anastomotic leakage. Ileostomy closure was arranged 1–3 months after the operation for patients with a preventive ileostomy.

## Postoperative complications and pathology

The postoperative complications were categorized as per the Clavien-Dindo method. Mild and serious complications were determined if the Clavien-Dindo classification was ≤II or ≥III, respectively (22). Anastomotic leak was defined as fecal flow through the abdominal drainage tube or signs of peritonitis, and the presence of an anastomotic leak was confirmed by abdominal CT. The anastomotic leak was graded following the International Study Group of Rectal Cancer grading: grade A, no special treatment required; grade B, active treatment required without reoperation; grade C, operative treatment (23). Postoperative tumor TNM pathological staging followed the American Joint Committee on Cancer (AJCC) 8th edition. An involved circumferential resection margin (CRM) was defined as ≤1 mm between the margin of deepest tumor infiltration and the surgical resection margin.

## Postoperative adjuvant chemotherapy

Stage I patients were followed up regularly, stage II patients received adjuvant oral 5-FU-based chemotherapy (capecitabine),

and stage III patients received Xelox (capecitabine plus oxaliplatin).

## Functional evaluation

Low anterior resection syndrone (LARS) score was utilized to assess bowel function. LARS scoring questionnaire, consisting of five questions about liquid stool and flatus incontinence, stools clustering, bowel frequency, and fecal urgency, was scored from 0 to 42. The patients were classified as no LARS, minor LARS, and major LARS when the score was 0–20, 21–29, and 30–42 points, respectively (24). The international prostate symptom score (IPSS) for urinary function consisted of seven items: urgency, frequency, nocturia, weak stream, intermittency, incomplete emptying, and straining (25). IPSS was classified as mild, moderate, or severe when the score was 0–7, 8–19, or 20–35, respectively. Prior to and at 6 and 12 months after surgery, the patients were given a questionnaire. At 6 and 12 months after ileostomy closure, patients with ileostomies completed questionnaires to assess bowel function.

## Follow up

After surgery, all patients were followed up every six months for the first three years and then annually for three to five years.

TABLE 1 Patients' baseline and clinical characteristics.

| | High ligation ($n = 134$) | Low ligation ($n = 86$) | *P* value |
|---|---|---|---|
| Age (years) | 63.6 ± 6.9 | 65.1 ± 6.8 | 0.110 |
| Gender | | | 0.442 |
| Male | 71 (53.0%) | 41 (47.7%) | |
| Female | 63 (47.0%) | 45 (52.3%) | |
| BMI (kg/m2) | 24.7 ± 2.5 | 25.0 ± 2.1 | 0.242 |
| ASA | | | 0.648 |
| I | 47 (35.1%) | 25 (29.1%) | |
| II | 69 (51.5%) | 48 (55.8%) | |
| III | 18 (13.4%) | 13 (15.1%) | |
| Tumor location | | | 0.302 |
| Upper rectum | 91 (67.9%) | 64 (74.4%) | |
| Lower rectum | 43 (32.1%) | 22 (25.6) | |
| Neoadjuvant therapy | 27 (18.8%) | 18 (20.9%) | 0.687 |
| History of abdominal surgery | 39 (29.1%) | 17 (19.8%) | 0.121 |
| Diabetes | 22 (16.4%) | 13 (15.1%) | 0.797 |
| Coronary heart disease | 27 (20.1%) | 18 (20.9%) | 0.889 |
| Hypertension | 31 (23.1%) | 26 (30.2%) | 0.241 |

Follow-up visits, conducted in the clinic and by telephone, included a physical examination, carcinoembryonic antigen measurement, CT of the chest and abdomen, colonoscopy, and completion of a questionnaire. If patients were found to suffer recurrent metastasis for the follow-up period, the location and point in time of recurrent metastasis were recorded. Patients with recurrent metastasis were reexamined every three months, assessing serum carcinoembryonic antigen and CT of the chest and abdomen.

## Statistical analysis

The study statistical analyses were done using macOS IBM SPSS Statistics 26.0. Comparing categorical variables and

TABLE 2 Surgical data and postoperative complications.

| | High ligation ($n = 134$) | Low ligation ($n = 86$) | *P* value |
|---|---|---|---|
| Conversion to open surgery | 5 (3.7%) | 2 (2.3%) | 0.708* |
| Operative time (min) | 184.6 ± 14.4 | 190.7 ± 16.4 | 0.004 |
| Intraoperative blood loss (ml) | 84.3 ± 24.5 | 91.2 ± 21.8 | 0.037 |
| Splenic flexure mobilization | 20 (14.9%) | 18 (20.9%) | 0.250 |
| Preventive ileostomy | 17 (12.7%) | 13 (15.1%) | 0.608 |
| Indwelling anal canal | 94 (70.1%) | 61 (70.9%) | 0.695 |
| Time to first flatus (day) | 3.6 ± 1.0 | 3.6 ± 1.1 | 0.539 |
| Hospital stays (day) | 10.2 ± 2.6 | 10.0 ± 3.0 | 0.720 |
| Postoperation complications | 34 (25.4%) | 18 (20.9%) | 0.449 |
| Dindo-Clavien classification | | | 0.734 |
| Mild | 23 (17.2%) | 13 (15.1%) | |
| Severe | 11 (8.2%) | 5 (5.8%) | |
| Incision infection | 5 (3.7%) | 4 (4.7%) | 0.739* |
| Intestinal obstruction | 3 (2.2%) | 1 (1.2%) | 1* |
| Diarrhea | 2 (1.5%) | 1 (1.2%) | 1* |
| Urinary retention | 5 (3.7%) | 2 (2.3%) | 0.708* |
| Pneumonia | 4 (3.0%) | 2 (2.3%) | 1* |
| Anastomotic bleeding | 1 (0.7%) | 1 (1.2%) | 1* |
| Anastomotic leakage | 14 (10.4%) | 7 (8.1%) | 0.570 |
| Leakage grade | | | 0.704* |
| A | 2 (1.5%) | 2 (2.3%) | |
| B | 5 (3.7%) | 2 (2.3%) | |
| C | 7 (5.2%) | 3 (3.5%) | |
| Reoperation | 8 (3.7%) | 3 (3.5%) | 0.534* |
| Overall 30-day mortality | 2 (1.5%) | 1 (1.2%) | 1* |

*Refers to Fisher's exact test.

continuous data among different groups was done *via* Chi-square or Fisher's exact tests. Relying on the distribution, the continuous data were evaluated with an independent *t*-test or Mann–Whitney *U* test. Univariate and multivariate logistic regression assessed the risk factors for anastomotic leakage. $p < 0.100$ variables were included in the multivariable analysis. The 5-year OS and DFS were analyzed by Kaplan-Meier curves, and, to verify the groups' significant differences, a log-rank test was done. $p < 0.05$ was regarded as statistically significant.

# Results

## Patient characteristics

Table 1 shows all patients' baseline and clinical characteristics. No statistically significant differences were found between patients who were treated with high ligation and those who underwent low ligation for gender, age, ASA stage, BMI, tumor location, neoadjuvant chemoradiotherapy, history of abdominal surgery, diabetes, coronary heart disease, and hypertension between the two groups ($p > 0.05$).

## Surgical data and postoperative complications outcomes

Table 2 shows the surgical outcomes and complications. The high ligation group had a shorter operation time than the low ligation group (184.6 ± 14.4 min vs. 190.7 ± 16.4 min, $p = 0.004$). Intraoperative blood loss in the high ligation group was significantly higher than in the low ligation group (91.2 ± 21.8 ml vs. 84.3 ± 24.5 ml, $p = 0.037$). No statistical differences were shown in conversion to open surgery, splenic flexure mobilization, preventive ileostomy, indwelling anal canal, time to first flatus, and hospital stay ($p > 0.05$). The incidence of postoperative complications in the high and low ligation groups was 25.4% and 20.9%, respectively, and no significant difference was observed ($p = 0.449$). The anastomotic leakage in the high and low ligation groups was 10.4% (14 patients) and 8.1% (7 patients), respectively, with no significant difference ($p = 0.570$). Reoperation occurred in the high and low ligation groups at 3.7% and 3.5%, respectively ($p = 0.534$). The 30-day after surgery mortality in the high and low ligation groups were two and one case, respectively, which was not significantly different.

## Anastomotic leakage risk factors

The male gender, neoadjuvant therapy, and the lower rectum tumors were considerably related to anastomotic leakage incidence, as revealed by univariable analysis. The male gender and the lower rectum tumors were considered anastomotic leakage risk factors, as the multivariable analysis revealed (Table 3).

## Pathological outcomes

Table 4 lists the pathological results summary. The tumor size, proximal margin, and distal margin were measured

TABLE 3 Risk factors for anastomotic leakage.

| | Anastomotic leakage | | Univariable analysis | | Multivariable analysis | |
|---|---|---|---|---|---|---|
| | Yes ($n = 21$) | No ($n = 199$) | *P* | OR (95%CI) | *P* | OR (95%CI) |
| **Gender** | | | | | | |
| Male | 17 | 96 | 0.011 | 4.333 (1.399–13.418) | 0.035 | 3.451 (1.091–10.919) |
| Female | 4 | 104 | | 1 (reference) | | 1 (reference) |
| **Age** | | | | | | |
| >65 | 11 | 84 | 0.492 | 1.381 (0.550–3.467) | – | – |
| <65 | 10 | 116 | | 1 (reference) | – | – |
| **Neoadjuvant therapy** | | | | | | |
| Yes | 8 | 38 | 0.038 | 2.296 (0.858–6.143) | 0.129 | 1.651 (0.599–4.545) |
| No | 13 | 162 | | 1 (reference) | | 1 (reference) |
| **Tumor location** | | | | | | |
| Lower rectum | 12 | 54 | 0.012 | 3.305 (1.298–8.414) | 0.047 | 2.628 (1.011–6.828) |
| Upper rectum | 9 | 146 | | 1 (reference) | | 1 (reference) |
| **Diverting ileostomy** | | | | | | |
| Yes | 4 | 27 | 0.852 | 0.884 (0.310–4.119) | – | – |
| No | 17 | 173 | | 1 (reference) | – | – |

without differences between the two groups ($p > 0.05$). The number of lymph nodes harvested in the high and low ligation groups was 16.3 ± 2.8 and 15.5 ± 2.4, respectively ($p = 0.053$). No significant difference was observed between the two groups in the number of positive lymph nodes ($p = 0.493$). No statistical differences were identified between the two groups in CRM, neural invasion, vascular invasion, degree of differentiation, pN stage, pT stage, and pTNM stage ($p > 0.05$; Table 4).

## Functional outcomes of LARS and IPSS questionnaires

The functional outcomes of LARS and IPSS questionnaires are shown in Table 5. No significant differences were observed before surgery, or 6 and 12 months following surgery, in both LARS and IPSS questionnaire responses between groups.

TABLE 4 Pathological data.

| | High ligation ($n = 134$) | Low ligation ($n = 86$) | *P* value |
|---|---|---|---|
| Tumor size (cm) | 3.8 ± 1.4 | 3.6 ± 1.5 | 0.170 |
| Proximal margin (cm) | 9.5 ± 2.2 | 8.9 ± 2.5 | 0.064 |
| Distal margin (cm) | 2.2 ± 1.1 | 1.9 ± 1.1 | 0.141 |
| CRM | | | 0.985 |
| Negative | 123 (91.8%) | 79 (91.9%) | |
| Positive | 11 (8.2%) | 7 (8.1%) | |
| Neural invasion | 21 (15.7%) | 10 (11.6%) | 0.400 |
| Vasculature invasion | 18 (13.4%) | 11 (12.8%) | 0.891 |
| Degree of differentiation | | | 0.534 |
| High | 95 (70.9%) | 55 (64.0%) | |
| Medium | 24 (17.9%) | 18 (20.9%) | |
| Low | 15 (11.2%) | 13 (15.1%) | |
| pT stage | | | |
| T1 | 8 (6.0%) | 6 (7.0%) | 0.612 |
| T2 | 19 (14.2%) | 13 (15.1%) | |
| T3 | 58 (43.3%) | 43 (50.0%) | |
| T4 | 49 (36.6%) | 24 (27.9%) | |
| pN stage | | | 0.749 |
| N0 | 96 (71.6%) | 65 (75.6%) | |
| N1 | 29 (21.6%) | 15 (17.4%) | |
| N2 | 9 (6.7%) | 6 (7.0%) | |
| pTNM | | | 0.803 |
| I | 27 (20.1%) | 19 (22.1%) | |
| II | 69 (51.5%) | 46 (53.5%) | |
| III | 38 (28.4%) | 21 (24.4%) | |
| Total number of lymph nodes harvested | 16.3 ± 2.8 | 15.5 ± 2.4 | 0.053 |
| Positive number of lymph nodes harvested | 0.8 ± 2.0 | 1.0 ± 2.2 | 0.493 |

## Long-time oncologic prognosis

The follow-up rate at five-years was 90.5%, with 12 patients in the high ligation group and nine patients in the low ligation lost to follow-up. The occurrence of recurrent metastases in the high and low ligation groups was 26.5% vs. 22.4%, respectively, which was not statistically different (Table 6). The 5-year OS

TABLE 5 Function outcomes of LARS and IPSS.

| | High ligation | Low ligation | *P* value |
|---|---|---|---|
| Preoperational LARS grade | 134 | 86 | 0.275 |
| No | 72 (53.7%) | 49 (64.5%) | |
| Minor | 45 (33.6%) | 18 (23.7%) | |
| Major | 17 (12.7%) | 9 (11.8%) | |
| 6-month LARS grade | 115 | 72 | 0.689 |
| No | 23 (20.0%) | 12 (16.7%) | |
| Minor | 55 (47.8%) | 39 (54.2%) | |
| Major | 37 (32.2%) | 21 (29.2%) | |
| 12-month LARS grade | 93 | 64 | 0.706 |
| No | 32 (34.4%) | 18 (28.1%) | |
| Minor | 46 (49.5%) | 35 (54.7%) | |
| Major | 15 (16.1%) | 11 (17.2%) | |
| Preoperational IPSS grade | 134 | 86 | 0.435 |
| Mild | 51 (38.1%) | 40 (46.5%) | |
| Moderate | 46 (34.3%) | 24 (27.9%) | |
| Sever | 37 (27.6%) | 22 (25.6%) | |
| 6-month IPSS grade | 115 | 72 | 0.699 |
| Mild | 27 (23.5%) | 19 (23.2%) | |
| Moderate | 47 (40.9%) | 38 (46.3%) | |
| Sever | 41 (35.7%) | 25 (30.5%) | |
| 12-month IPSS grade | 93 | 64 | 0.171 |
| Mild | 31 (33.3%) | 17 (26.6%) | |
| Moderate | 34 (36.6%) | 33 (51.6%) | |
| Sever | 28 (30.1%) | 14 (21.9%) | |

TABLE 6 Recurrent metastasis and long-time outcome.

| | High ligation ($n = 132$) | Low ligation ($n = 85$) | *P* value |
|---|---|---|---|
| Recurrent metastasis | 35 (26.5%) | 19 (22.4%) | 0.489 |
| Liver metastasis | 15 (11.4%) | 8 (9.4%) | 0.648 |
| Pulmonary metastasis | 10 (7.6%) | 3 (3.5%) | 0.220 |
| Liver and pulmonary metastasis | 8 (6.1%) | 7 (8.2%) | 0.538 |
| Local recurrence | 2 (1.5%) | 1 (1.2%) | 1* |
| 5-year overall survival | 78.3% | 82.4% | 0.463 |
| 5-year disease-free survival | 72.4% | 76.6% | 0.485 |

*Refers to Fisher's exact test.

and DFS for the high and low ligation groups were 78.3% vs. 82.4% ($p = 0.463$), and 72.4% vs. 76.6% ($p = 0.485$), respectively, showing no statistical differences (Figures 1 and 2).

## Discussion

This study discloses that low ligation increases operation time and intraoperative blood loss compared to high ligation. Furthermore, no significant difference was observed between the high and low ligation groups in anastomotic leakage. The male gender and lower rectum tumors are anastomotic leakage risk factors. No significant differences were observed between the groups in oncologic outcomes, such as 5-year OS and 5-year DFS, as well as functional outcomes, such as bowel and urine functions.

The study revealed that the low ligation group had a longer operative time (184.56 ± 14.4 vs. 190.7 ± 16.4, $p = 0.004$) and more intraoperative blood loss (84.3 ± 24.5 vs. 91.2 ± 21.8, $p = 0.037$). Given that low-ligation works by lymph node clearance at IMA root to expose the left colic artery, on the premise of IMA safety, it is harder to run and requires more experienced surgeons. A recent study found that whether the left colic artery is preserved or not was independent of the operative time and intra-operative blood loss (5). Similarly, some meta-analyses showed that the left colic artery preservation would not increase the operative time and intra-operative blood loss (26, 27). However, Park et al. (8) reported that the low-ligation strategy contributed to a shorter operative time but was not superior in decreasing intraoperative blood loss. The discrepancy might be associated with the operative experience of the surgeons.

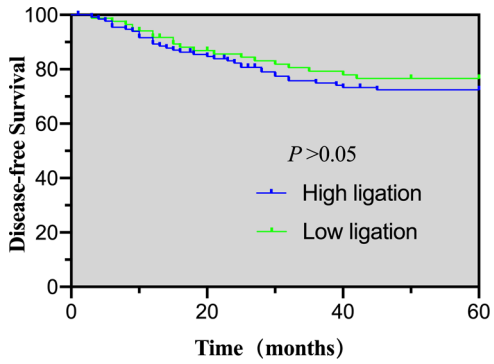


FIGURE 1
5-year disease-free survival of the high- and low-ligation groups.

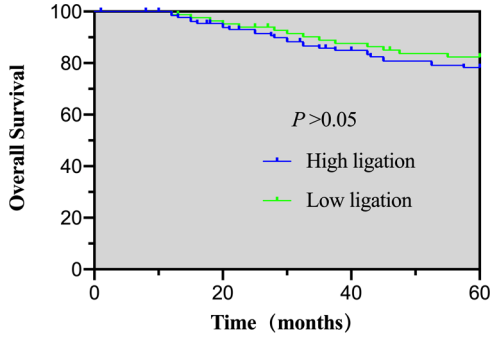


FIGURE 2
5-year overall survival of the high- and low-ligation groups.

Anastomotic leakage is a high-risk complication during low anterior rectal resections, leading to a longer hospital stay and higher medical costs, as well as increasing the ileostomy rate and mortality. In the current research, the anastomotic leakage incidence in the high and low ligation groups were 10.4% and 8.1%, respectively, which was not statistically significant, and the male gender and the lower rectum tumor were considered risk factors. Several studies showed that the anastomotic leakage incidence during low anterior rectal resections would not be increased when high-ligation was applied, and gender and the distance from the tumor to the anus were major factors causing anastomotic leakage after operation (28, 29). The level of IMA ligation does not correlate with anastomotic leakage and must be selected according to factors, including the presence or absence of tension anastomosis (30). Additionally, it has been suggested that anatomical variants of the left colic artery should be of concern, as insufficient vascularization of the proximal colonic conduit in the absence of the left colic artery is also an important factor in the occurrence of anastomotic leakage (31). However, anastomotic blood perfusion remains one of the important factors affecting anastomotic healing. Intraoperative colonic perfusion was measured using laser Doppler flowmetry and was found to be slightly decreased in the high ligation group and slightly increased in the low ligation group, independent of blood pressure (16). Seike et al. (15) reported that, after clamping the IMA, the anastomotic blood flow of the proximal colon was significantly reduced, which was more evident in elderly men, along with a higher risk of anastomotic leakage. Other studies also demonstrated that low ligation could decrease the anastomotic leakage risk (32–34). Therefore, larger samples are required to further explore the relationship between anastomotic leakage and IMA ligation level in the future.

Bowel and urinary function were poor after rectal cancer surgery. A Japanese randomized controlled trial reported no significant differences between patients with high and low ligation, assessed at three months and one year postoperatively, on defecatory function, fecal incontinence quality of life scale defaecation self-assessment, or continence score (9). Defecation function, related to levels of IMA ligation resulting in different blood supply to the anastomosis, is also related to other factors, such as the denervated neorectum motility, rectal compliance, anal sphincter, and anastomosis level. Although neither group returned to preoperative IPSS levels, there was an improvement in IPSS at nine months after low ligation compared to high

ligation (20). Park (8) reported no difference in bowel and urinary function between the two groups before surgery, three months after surgery, and 12 months after surgery. Similarly, this research revealed no significant statistical difference in LARS and IPSS between high and low ligation in preoperative, six months postoperative, and 12 months postoperative. The difference may be related to autonomic nerve injury during IMA peripheral lymph node dissection in our surgery.

The number of lymph nodes dissected during operation is vital for operation assessment, guiding postoperative adjuvant chemoradiotherapy and prognosis. Patients with lymph node metastasis are more likely to have tumor recurrence and experience a shorter survival time compared to those without metastasis (35). Research revealed no evidence showing the benefits of high-ligation in long-term prognosis, although it could get more lymph nodes dissected (36). Many current studies have suggested that there were no more lymph nodes dissected by high-ligation, and still, no superiority was demonstrated in long-term prognosis compared to low-ligation (37–41). There were no statistical differences between the total number of lymph nodes dissected and the number of positive ones. Moreover, the 5-year OS and 5-year DFS showed no evident differences between the two groups.

Several limitations remain in this study. First, this is a retrospective study involving a small sample size from a single institute, requiring larger-scale, multi-center, and randomized controlled trials for further validation. Second, selection bias might not be ignored. Finally, sexual functions, such as the international index of erectile function (IIEF-5), and the female sexual function index (FSFI), were not assessed in this study.

## Conclusions

The ligation level of IMA has no significant effect on the incidence of anastomotic leakage, defecation, urinary function, or long-term prognosis. However, larger randomized controlled trials are still required to further validate this result.

## Data availability statement

The raw data supporting the conclusions of this article will be made available by the authors, without undue reservation.

## Ethics statement

The studies involving human participants were reviewed and approved by the Ethics Committee of the Second Affiliated Hospital of Chongqing Medical University. The patients/participants provided their written informed consent to participate in this study. Written informed consent was obtained from the individual(s) for the publication of any potentially identifiable images or data included in this article.

## Author contributions

All authors contributed to the study conception and design. Material preparation, data collection and analysis were performed by YC and PZ. The first draft of the manuscript was written by JY. LT, CL and BS prepared **Tables 1–6**. ZZ and QY prepared Figures 1–2. JZ commented on previous versions of the manuscript. All authors contributed to the article and approved the submitted version.

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## Conflict of interest

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

## Publisher's note



## References

1. Siegel RL, Miller KD, Fuchs HE, Jemal A. Cancer statistics, 2021. *CA Cancer J Clin.* (2021) 71:7–33. doi: 10.3322/caac.21654

2. Cirocchi R, Trastulli S, Farinella E, Desiderio J, Vettoretto N, Parisi A, et al. High tie versus low tie of the inferior mesenteric artery in colorectal cancer: a RCT is needed. *Surg Oncol.* (2012) 21:e111–123. doi: 10.1016/j.suronc.2012.04.004

3. Dimitriou N, Felekouras E, Karavokyros I, Pikoulis E, Vergadis C, Nonni A, et al. High versus low ligation of inferior mesenteric vessels in rectal cancer surgery: a retrospective cohort study. *J Buon.* (2018) 23:1350–61. PMID: 30570858

4. Fujii S, Ishibe A, Ota M, Suwa H, Watanabe J, Kunisaki C, et al. Short-term and long-term results of a randomized study comparing high tie and low tie inferior mesenteric artery ligation in laparoscopic rectal anterior resection:

subanalysis of the HTLT (high tie vs. Low tie) study. *Surg Endosc*. (2019) 33:1100–10. doi: 10.1007/s00464-018-6363-1

5. Chen JN, Liu Z, Wang ZJ, Zhao FQ, Wei FZ, Mei SW, et al. Low ligation has a lower anastomotic leakage rate after rectal cancer surgery. *World J Gastrointest Oncol*. (2020) 12:632–41. doi: 10.4251/wjgo.v12.i6.632

6. Hajibandeh S, Hajibandeh S, Maw A. Meta-analysis and trial sequential analysis of randomized controlled trials comparing high and low ligation of the Inferior mesenteric artery in rectal cancer surgery. *Dis Colon Rectum*. (2020) 63:988–99. doi: 10.1097/dcr.0000000000001693

7. Guo Y, Wang D, He L, Zhang Y, Zhao S, Zhang L, et al. Marginal artery stump pressure in left colic artery-preserving rectal cancer surgery: a clinical trial. *ANZ J Surg*. (2017) 87:576–81. doi: 10.1111/ans.13032

8. Park SS, Park B, Park EY, Park SC, Kim MJ, Sohn DK, et al. Outcomes of high versus low ligation of the inferior mesenteric artery with lymph node dissection for distal sigmoid colon or rectal cancer. *Surg Today*. (2020) 50:560–8. doi: 10.1007/s00595-019-01942-2

9. Matsuda K, Hotta T, Takifuji K, Yokoyama S, Oku Y, Watanabe T, et al. Randomized clinical trial of defaecatory function after anterior resection for rectal cancer with high versus low ligation of the inferior mesenteric artery. *Br J Surg*. (2015) 102:501–8. doi: 10.1002/bjs.9739

10. Girard E, Trilling B, Rabattu PY, Sage PY, Taton N, Robert Y, et al. Level of inferior mesenteric artery ligation in low rectal cancer surgery: high tie preferred over low tie. *Tech Coloproctol*. (2019) 23:267–71. doi: 10.1007/s10151-019-01931-0

11. Bonnet S, Berger A, Hentati N, Abid B, Chevallier JM, Wind P, et al. High tie versus low tie vascular ligation of the inferior mesenteric artery in colorectal cancer surgery: impact on the gain in colon length and implications on the feasibility of anastomoses. *Dis Colon Rectum*. (2012) 55:515–21. doi: 10.1097/DCR.0b013e318246f1a2

12. Thum-umnuaysuk S, Boonyapibal A, Geng YY, Pattana-Arun J. Lengthening of the colon for low rectal anastomosis in a cadaveric study: how much can we gain? *Tech Coloproctol*. (2013) 17:377–81. doi: 10.1007/s10151-012-0930-6

13. Charan I, Kapoor A, Singhal MK, Jagawat N, Bhavsar D, Jain V, et al. High ligation of Inferior mesenteric artery in left colonic and rectal cancers: lymph node yield and survival benefit. *Indian J Surg*. (2015) 77:1103–8. doi: 10.1007/s12262-014-1179-2

14. Park MG, Hur H, Min BS, Lee KY, Kim NK. Colonic ischemia following surgery for sigmoid colon and rectal cancer: a study of 10 cases and a review of the literature. *Int J Colorectal Dis*. (2012) 27:671–5. doi: 10.1007/s00384-011-1372-8

15. Seike K, Koda K, Saito N, Oda K, Kosugi C, Shimizu K, et al. Laser Doppler assessment of the influence of division at the root of the inferior mesenteric artery on anastomotic blood flow in rectosigmoid cancer surgery. *Int J Colorectal Dis*. (2007) 22:689–97. doi: 10.1007/s00384-006-0221-7

16. Komen N, Slieker J, de Kort P, de Wilt JH, van der Harst E, Coene PP, et al. High tie versus low tie in rectal surgery: comparison of anastomotic perfusion. *Int J Colorectal Dis*. (2011) 26:1075–8. doi: 10.1007/s00384-011-1188-6

17. Sörelius K, Svensson J, Matthiessen P, Rutegård J, Rutegård M. A nationwide study on the incidence of mesenteric ischaemia after surgery for rectal cancer demonstrates an association with high arterial ligation. *Colorectal Dis*. (2019) 21:925–31. doi: 10.1111/codi.14674

18. Fiori E, Crocetti D, Lamazza A, Felice FD, Sterpetti AV, Irace L, et al. Is low Inferior mesenteric artery ligation worthwhile to prevent urinary and sexual dysfunction after total mesorectal excision for rectal cancer? *Anticancer Res*. (2020) 40:4223–8. doi: 10.21873/anticanres.14423

19. You X, Liu Q, Wu J, Wang Y, Huang C, Cao G, et al. High versus low ligation of inferior mesenteric artery during laparoscopic radical resection of rectal cancer: a retrospective cohort study. *Medicine*. (2020)) 99:e19437. doi: 10.1097/md.0000000000019437

20. Mari GM, Crippa J, Cocozza E, Berselli M, Livraghi L, Carzaniga P, et al. Low ligation of inferior mesenteric artery in laparoscopic anterior resection for rectal cancer reduces genitourinary dysfunction: results from a randomized controlled trial (HIGHLOW trial). *Ann Surg*. (2019) 269:1018–24. doi: 10.1097/sla.0000000000002947

21. Kverneng Hultberg D, Afshar AA, Rutegård J, Lange M, Haapamäki MM, Matthiessen P, et al. Level of vascular tie and its effect on functional outcome 2 years after anterior resection for rectal cancer. *Colorectal Dis*. (2017) 19:987–95. doi: 10.1111/codi.13745

22. Clavien P, Barkun J, de Oliveira M, Vauthey J, Dindo D, Schulick R, et al. The clavien-dindo classification of surgical complications: five-year experience. *Ann Surg*. (2009) 250:187–96. doi: 10.1097/SLA.0b013e3181b13ca2

23. Rahbari N, Weitz J, Hohenberger W, Heald R, Moran B, Ulrich A, et al. Definition and grading of anastomotic leakage following anterior resection of the rectum: a proposal by the international study group of rectal cancer. *Surgery*. (2010) 147:339–51. doi: 10.1016/j.surg.2009.10.012

24. Emmertsen KJ, Laurberg S. Low anterior resection syndrome score: development and validation of a symptom-based scoring system for bowel dysfunction after low anterior resection for rectal cancer. *Ann Surg*. (2012) 255:922–8. doi: 10.1097/SLA.0b013e31824f1c21

25. Barry MJ, Fowler Jr. FJ, O'Leary M P, Bruskewitz RC, Holtgrewe HL, Mebust WK, et al. The American urological association symptom index for benign prostatic hyperplasia. *J Urol*. (2017) 197:S189–97. doi: 10.1016/j.juro.2016.10.071

26. Liu J, Gong Y, He M, Zeng X, Liu Y. Clinical effect of preservation or nonpreservation of left colic artery in total mesorectal excision under laparoscopy: a meta-analysis. *Gastroenterol Res Pract*. (2020) 2020:1958573. doi: 10.1155/2020/1958573

27. Fan YC, Ning FL, Zhang CD, Dai DQ. Preservation versus non-preservation of left colic artery in sigmoid and rectal cancer surgery: a meta-analysis. *Int J Surg*. (2018) 52:269–77. doi: 10.1016/j.ijsu.2018.02.054

28. Kong M, Chen H, Xin Y, Jiang Y, Han Y, Sheng H. High ligation of the inferior mesenteric artery and anastomotic leakage in anterior resection for rectal cancer: a systematic review and meta-analysis of randomized controlled trial studies. *Colorectal Dis*. (2021) 23:614–24. doi: 10.1111/codi.15419

29. Fujii S, Ishibe A, Ota M, Watanabe K, Watanabe J, Kunisaki C, et al. Randomized clinical trial of high versus low inferior mesenteric artery ligation during anterior resection for rectal cancer. *BJS Open*. (2018) 2:195–202. doi: 10.1002/bjs5.71

30. Draginov A, Chesney TR, Quereshy HA, Chadi SA, Quereshy FA. Association of high ligation versus low ligation of the inferior mesenteric artery on anastomotic leak, postoperative complications, and mortality after minimally invasive surgery for distal sigmoid and rectal cancer. *Surg Endosc*. (2020) 34:4593–600. doi: 10.1007/s00464-019-07203-0

31. Cirocchi R, Randolph J, Cheruiyot I, Davies JR, Wheeler J, Lancia M, et al. Systematic review and meta-analysis of the anatomical variants of the left colic artery. *Colorectal Dis*. (2020) 22:768–78. doi: 10.1111/codi.14891

32. Si MB, Yan PJ, Du ZY, Li LY, Tian HW, Jiang WJ, et al. Lymph node yield, survival benefit, and safety of high and low ligation of the inferior mesenteric artery in colorectal cancer surgery: a systematic review and meta-analysis. *Int J Colorectal Dis*. (2019) 34:947–62. doi: 10.1007/s00384-019-03291-5

33. Zeng J, Su G. High ligation of the inferior mesenteric artery during sigmoid colon and rectal cancer surgery increases the risk of anastomotic leakage: a meta-analysis. *World J Surg Oncol*. (2018) 16:157. doi: 10.1186/s12957-018-1458-7

34. Luo Y, Yu MH, Huang YZ, Jing R, Qin J, Qin SL, et al. Lymphadenectomy around inferior mesenteric artery in low-tie vs high-tie laparoscopic anterior resection: short- and long-term outcome of a cohort of 614 rectal cancers. *Cancer Manag Res*. (2021) 13:3963–71. doi: 10.2147/cmar.S282986

35. Zhao X, Ma J, Fu Z, Hong H, Zhang L, Xue P, et al. Prognostic value of apical lymph node metastasis at the inferior mesenteric artery in sigmoid and rectal cancer patients who undergo laparoscopic surgery. *J Surg Oncol*. (2021) 123(Suppl 1):S88–s94. doi: 10.1002/jso.26346

36. Titu LV, Tweedle E, Rooney PS. High tie of the inferior mesenteric artery in curative surgery for left colonic and rectal cancers: a systematic review. *Dig Surg*. (2008) 25:148–57. doi: 10.1159/000128172

37. Matsuda K, Yokoyama S, Hotta T, Takifuji K, Watanabe T, Tamura K, et al. Oncological outcomes following rectal cancer surgery with high or low ligation of the inferior mesenteric artery. *Gastrointest Tumors*. (2017) 4:45–52. doi: 10.1159/000477805

38. Nayeri M, Iskander O, Tabchouri N, Artus A, Michot N, Muller O, et al. Low tie compared to high tie vascular ligation of the inferior mesenteric artery in rectal cancer surgery decreases postoperative complications without affecting overall survival. *Anticancer Res*. (2019) 39:4363–70. doi: 10.21873/anticanres.13605

39. Yasuda K, Kawai K, Ishihara S, Murono K, Otani K, Nishikawa T, et al. Level of arterial ligation in sigmoid colon and rectal cancer surgery. *World J Surg Oncol*. (2016) 14:99. doi: 10.1186/s12957-016-0819-3

40. Zhang C, Chen L, Cui M, Xing J, Yang H, Yao Z, et al. Short- and long-term outcomes of rectal cancer patients with high or improved low ligation of the inferior mesenteric artery. *Sci Rep*. (2020) 10:15339. doi: 10.1038/s41598-020-72303-0

41. Luo Y, Li R, Wu D, Zeng J, Wang J, Chen X, et al. Long-term oncological outcomes of low anterior resection for rectal cancer with and without preservation of the left colic artery: a retrospective cohort study. *BMC Cancer*. (2021) 21:171. doi: 10.1186/s12885-021-07848-y

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# Concomitant preoperative airflow obstruction confers worse prognosis after trans-thoracic surgery for esophageal cancer

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**Background:** Airflow obstruction is a critical element of chronic airway diseases. This study aimed to evaluate the impact of preoperative airflow obstruction on the prognosis of patients following surgery for esophageal carcinoma.

**Methods:** A total of 821 esophageal cancer patients were included and classified into two groups based on whether or not they had preoperative airflow obstruction. Airflow obstruction was defined as a forced expiration volume in the first second ($FEV_1$)/forced vital capacity (FVC) ratio below the lower limit of normal (LLN). A retrospective analysis of the impact of airflow obstruction on the survival of patients with esophageal carcinoma undergoing esophagectomy was performed.

**Results:** Patients with airflow obstruction (102/821, 12.4%) had lower three-year overall (42/102, 58.8%) and progression-free survival rate (47/102, 53.9%) than those without airflow obstruction ($P < 0.001$). Multivariate analyses showed that airflow obstruction was an independent risk factor for overall survival (Hazard Ratio = 1.66; 95% CI: 1.17–2.35, $P = 0.004$) and disease progression (Hazard Ratio = 1.51; 95% CI: 1.1–2.08; $P = 0.01$). A subgroup analysis revealed that the above results were more significant in male patients, BMI < 23 kg/m$^2$ patients or late-stage cancer (stage III-IVA) ($P = 0.001$) patients and those undergoing open esophagectomy ($P < 0.001$).

**Conclusion:** Preoperative airflow obstruction defined by $FEV_1$/FVC ratio below LLN was an independent risk factor for mortality in esophageal cancer patients after trans-thoracic esophagectomy. Comprehensive management of airflow obstruction and more personalized surgical decision-making are necessary to improve survival outcomes in esophageal cancer patients.

KEYWORDS

esophageal cancer, survival, airflow obstruction, lung function, decision-making

Abbreviations

EC, esophageal cancer; AFO, airflow obstruction; LLN, lower limit of normal; $FEV_1$, forced expiration volume in the first second; FVC, forced vital capacity; OS, overall survival; PFS, progression-free survival; G, grade of tumor differentiation; pT, pathological T factor; pN, pathological N factor; BMI, body mass index; MIE, minimally invasive esophagectomy.

# Introduction

Esophageal cancer (EC) is a highly aggressive malignancy with an inferior prognosis of 5-year survival rate of about 20% over the past decade worldwide (1). The incidence and healthcare burden of esophageal cancer in Eastern Asia were higher than in the rest of the world over the past decades (2). Though esophagectomy is an essential treatment for esophageal cancer, it is associated with a high incidence of postoperative complications (3, 4), and overall outcomes are still poor for late-stage esophageal cancer, especially in squamous cell cancer (5, 6).

Lung function is a criterion for eligibility for radical esophagectomy (7). Esophageal cancer patients undergoing esophagectomy should have good or at least not poor lung function, as many patients with severe chronic pulmonary disease are unsuitable for thoracic surgery. It is widely accepted that smoking is one of the relevant risk factors for esophageal cancer and chronic obstructive airway disease (8). Previous research demonstrated a high degree of overlap (7.1%–25%) of operable esophageal cancer patients with chronic obstructive pulmonary diseases (COPD) or asthma (9–11). Furthermore, chronic airway obstruction is directly related to the morbidity of esophagectomy, particularly concerning pulmonary complications and anastomotic leaks (10, 11). However, studies on the outcomes of patients with esophageal cancer and COPD or asthma were limited to postoperative morbidity rather than survival status. Though preoperative low vital capacity decreased the survival rate after radical esophagectomy for cancer (12), the impact of preoperative airway obstruction on long-term survival is unclear. Thus, an accurate assessment of the risk of airway obstruction in esophageal cancer patients is essential. We conducted this study to investigate the impact of preoperative airway obstruction on survival outcomes in patients with esophageal cancer after trans-thoracic esophagectomy. These findings shed light on patients' long-term airway management after esophageal cancer surgery.

# Materials and methods

## Population

This is a single-center, retrospective cohort study. From June 2012 to December 2015, 1,016 Chinese patients with esophageal cancer admitted to Zhongshan Hospitals, Fudan University (Shanghai, China), were evaluated and enrolled in the present study. All patients underwent radical trans-thoracic esophagectomy (Ivor-Lewis or McKeown procedure) with gastroesophageal reconstruction. Forty-four patients lost to follow-up, 149 patients without retrieved preoperative spirometry records, and two patients with distant metastasis at the time of diagnosis (M1) were excluded from the sample (**Figure 1**). This study was approved by the Ethics Committee of Zhongshan Hospital, Fudan University.

## Data collection for baseline patients' characteristics

Esophageal carcinoma and the stage were pathologically determined.

Information about patient characteristics and short-term postoperative complications before hospital discharge was obtained from the patient's medical records. Demographic characteristics, clinicopathological features, pulmonary function, and details of postoperative complications were collected and summarized in **Tables 1**, **2**.

## Evaluation of preoperative pulmonary function variables by spirometry

Spirometry was performed in Zhongshan hospital according to the ATS standards (13). Airflow obstruction was defined as a forced expiration volume in the first second (FEV1)/forced vital capacity (FVC) ratio was below the lower fifth percentile of a large healthy Chinese reference group (lower limit of normal, LLN) (14–16). The lower limit of normal (LLN) of FEV1/FVC was calculated with the formula in **Supplementary Table S1**. A website was developed by our team for convenient calculation and diagnosis of airflow obstruction (https://drpulmonary.shinyapps.io/AOdiagnosistool/). Given that FEV1/FVC decreases with increased age and most of the study population were over 50 years old, LLN definition of airflow obstruction was used to minimize false positives.

## Postoperative complications

Postoperative complications, including pulmonary complications (e.g., pneumonia, acute respiratory distress syndrome, and aspiration), anastomotic leakage, surgical site infection, cardiac complications, chyle leakage, thromboembolic events, recurrent laryngeal nerve paresis, and other complications were summarized. The severity of postoperative complications was classified according to the Clavien-Dindo classification as instructed by the International Consensus on Standardization of Data Collection for Complications Associated With Esophagectomy (17). Overall complications were defined as grade II and higher according to the Clavien-Dindo classification.

## Follow-Up and definition of recurrence

In principle, patients were reviewed through in-clinic follow-ups every three months in the first year and every six months

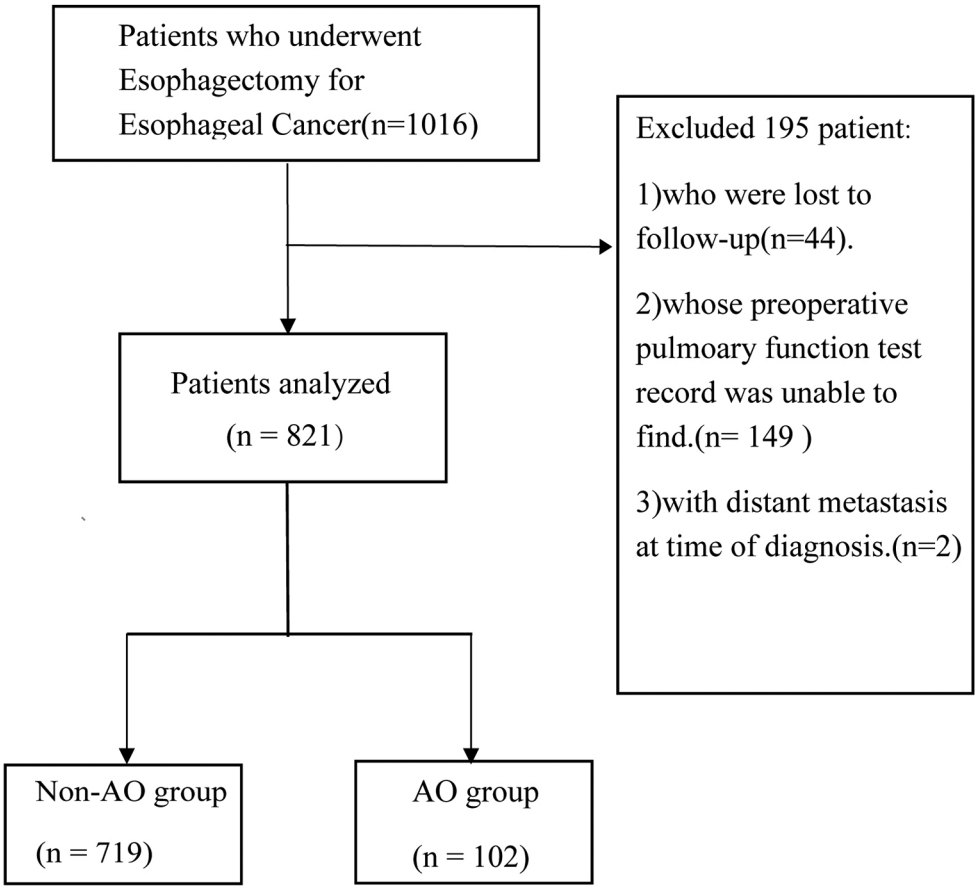


FIGURE 1
Study profile. Schematic diagram showing the study profile. The AFO group included patients with an $FEV_1$/FVC ratio below the LLN. AFO: airflow obstruction.

after that for at least 3 years. Computed tomography of the neck, chest, and abdomen was examined every six months. Disease progression was defined as local recurrence of primary esophageal cancer, distant metastasis, or death due to any cause.

## Statistical analysis

All collected data were manually checked for completeness and consistency, and the continuous variables were tested for normality using the Shapiro–Wilk test. Normally distributed variables were compared using the *t*-test, and non-normally distributed ones were compared using the Mann–Whitney *U* test between airflow obstruction and non-airflow obstruction groups. Comparisons between the proportions were made using the $\chi 2$ test or Fisher's exact test. Survival was calculated using Kaplan–Meier survival curves and compared using the log-rank test. $P < 0.05$ was considered significant. Median follow-up time was calculated using the reverse Kaplan–Meier method (18). The Cox proportional hazards model was used for the univariate and multivariate analyses to identify independent risk factors associated with survival. Risk-adjusted, restricted *cubic* splines with 4 knots were used to model the possible non-linearity of the association between BMI and the risk of all-cause death (19, 20). The R *Code* for restricted cubic splines analysis is available on the GitHub repository: https://github.com/longerham/RCS#rcs. Data analysis was performed using R Foundation Statistical software (R 3.2.2) with ggplot2, forest plot, and survival packages (The R Foundation for Statistical Computing, Vienna, Austria).

# Results

## Distribution of characteristics in the study population

Among included 821 patients with esophageal cancer, 102 patients were with airflow obstruction (FEV1/FVC < LLN, AFO group), and the remaining 719 patients were classified as

TABLE 1 Baseline demographics, clinicopathological and spirometric characteristics of Non-AFO and AFO patients.

| | Total, $n = 821$ | Non-airflow obstruction, $n = 719$ | Airflow obstruction, $n = 102$ | $p$ value |
|---|---|---|---|---|
| Age, year[#] | 61.2 (38–84) | 60.9 (38–84) | 62.5 (44–77) | 0.04* |
| Gender | | | | <0.001* |
| Male | 626 (76.2) | 525 (73) | 91 (89.2) | |
| Female | 195 (23.8) | 194 (27) | 11 (10.8) | |
| BMI, kg/m$^2$ | 22.8 (14.8–34.5) | 22.9 (14.8–34.5) | 22.2 (15.6–30.5) | 0.019* |
| Smoke | | | | 0.003* |
| Current or ever | 355 (43.3) | 297 (41.3) | 58 (56.9) | |
| Never | 466 (56.7) | 422 (58.7) | 44 (43.1) | |
| Pulmonary Function | | | | |
| $FEV_1$/FVC | 77.51 (45.39–99.08) | 79.57 (67.33–99.08) | 63.01 (45.39–69.7) | <0.001* |
| $FEV_1$, L | 2.58 (0.97–4.53) | 2.66 (1.08–4.53) | 2.06 (0.97–3.64) | <0.001* |
| FVC, L | 3.34 (1.09–5.74) | 3.35 (1.09–5.74) | 3.26 (1.54–5.22) | 0.26 |
| %FVC | 92.93 (27.81–132.36) | 93.55 (27.81–145.9) | 88.52 (51.2–132.3) | <0.001* |
| DLCO[1], mL/min mHg$^{-1}$ | 19.33 (1.42–32.67) | 19.58 (2.83–35.72) | 17.4 (1.42–32.67) | 0.002* |
| %DLCO[1] | 91.51 (13.19–175/96) | 92.93 (13.19–175.96) | 80.95 (16.51–147.95) | <0.001* |
| Neoadjuvant Chemotherapy | | | | 0.39 |
| Yes | 84 (10.2) | 78 (10.6) | 8 (7.8) | |
| No | 737 (89.8) | 643 (89.4) | 94 (92.2) | |
| Approach | | | | 0.72 |
| Open | 432 (52.6) | 380 (52.9) | 52 (50.98) | |
| MIE | 389 (47.4) | 339 (47.1) | 50 (49.02) | |
| pG | | | | 0.47 |
| G3 | 280 (34.1) | 242 (33.7) | 38 (37.4) | |
| G1-2 | 541 (65.9) | 477 (66.3) | 64 (62.6) | |
| pT | | | | 0.20 |
| T0-1 | 278 (33.9) | 250 (34.8) | 28 (27.5) | |
| T2 | 206 (25.1) | 178 (24.8) | 28 (27.5) | |
| T3-4 | 337 (41) | 291 (40.4) | 46 (45) | |
| pN | | | | 0.35 |
| N0 | 543 (66.1) | 483 (67.2) | 60 (58.9) | |
| N1 | 174 (21.2) | 146 (20.3) | 28 (27.4) | |
| N2-3 | 104 (12.7) | 90 (12.5) | 14 (13.7) | |
| Histology | | | | 0.018* |
| SCC | 768 (93.5) | 667 (92.8) | 101 (99.0) | |
| Others | 53 (6.5) | 52 (7.2) | 1 (1.0) | |
| Tumor length (cm)[#] | 3.19 (1–10) | 3.18 (1–10) | 3.25 (1–8) | 0.61 |
| Tumor location | | | | 0.76 |
| Upper | 73 (8.9) | 62 (8.6) | 11 (10.8) | |
| Middle | 486 (59.2) | 426 (59.3) | 60 (58.8) | |
| Lower | 262 (31.9) | 231 (32.2) | 31 (30.4) | |
| PNI | | | | 0.39 |
| Yes | 129 (15.8) | 110 (15.3) | 19 (18.6) | |
| No | 691 (84.2) | 608 (84.6) | 83 (81.4) | |
| LVSI | | | | 0.82 |
| Yes | 99 (12.1) | 86 (12) | 13 (12.7) | |
| No | 722 (87.9) | 633 (88) | 89 (87.3) | |

BMI, body mass index; FEV1/FVC, forced expiratory volume in 1s/vital capacity; FEV1, forced expiratory volume in 1s; FVC, forced vital capacity; %VC, %forced vital capacity; DLCO, diffusing capacity; MIE, Minimally invasive esophagectomy; pT, pathological T factor; pN, pathological N factor; pStage, pathological Stage. SCC, Squamous cell carcinoma; PNI, perineural invasion; LVSI, lymph-vascular space invasion.

*$p$ value < 0.05.

[#]Data are shown as median (range). All other data are shown as numbers (%) or mean (range).

[1]Missing data. DLCO and % DLCO were missing for 3.3% ($N = 27$).

TABLE 2 Operative outcomes among the study populations.

| | | Airflow Obstrcution, $n = 102$ | Non-airflow Obstruction, $n = 719$ | Total, $n = 821$ | $p$ value |
|---|---|---|---|---|---|
| **Median hospital stay (days)**[#] | **Preoperative** | 3 (1–13) | 3 (1–21) | 3 (1–21) | 0.42 |
| | **Postoperative** | 13 (7–105) | 12 (6–197) | 12 (6–197) | 0.11 |
| | **Total** | 18 (9–108) | 16 (8–200) | 16 (8-200) | 0.18 |
| **Overall complications (≥Grade II)** | | 50 (49) | 287 (39.9) | 337 (41) | 0.08 |
| **Anastomotic leakage** | | 28 (27.5) | 105 (14.6) | 133 (16.2) | 0.001* |
| **Pulmonary complications** | | 15 (14.7) | 129 (17.9) | 144 (17.5) | 0.42 |
| **Lung metastasis** | | 18 (17.65) | 58 (8.1) | 76 (8.9) | <0.001* |
| **Mediastinal lymph node metastasis** | | 10 (9.8) | 95 (13.2) | 105 (12.8) | 0.34 |

*$p$ value < 0.05.
[#]Data are shown as median (range). All other data are shown as numbers (%).

non-airflow obstruction ($FEV_1/FVC \geq LLN$, non-AFO group) patients. Table 1 showed that non-airflow obstruction patients were younger than airflow obstruction patients (mean 60.9 vs. 62.5 years; $P < 0.001$). Airflow obstruction was associated with male ($P < 0.001$), lower BMI (mean 22.2 vs. 22.9 kg/m$^2$; $P = 0.019$), smoking history ($P = 0.003$), and squamous cell carcinoma ($P = 0.018$) No significant differences in tumor grades (G), pathological T factor (pT); pathological N factor (pN), perineural invasion (PNI), lymph-vascular space invasion (LVSI), tumor length or tumor locations between two groups were discovered. Table 1 also demonstrated the differences in spirometric variables and operative procedures between AFO and non-AFO groups. $FEV_1/FVC$, $FEV_1$, %VC predicted, and DLCO variables in AFO group were significantly lower than those in non-AFO group.

## Short-term outcomes in AFO and non-AFO groups

Length of hospital stay and incidence of overall complications, pulmonary complications, and anastomotic leaks were given in Table 2. Airflow obstruction patients showed significantly higher rate of anastomotic leakage than non-airflow obstruction patients (27.5% vs. 14.6%; $P < 0.001$). However, there were no significant differences between the groups in the length of hospital stay and rates of pulmonary complications.

## Impact of airflow obstruction on survival of esophageal cancer patients

The median follow-up time was 54 months for all patients, while the median follow-up time was 53.6 months (95% CI: 51.9–56.1) in non-AFO group and 55.9 months (95% CI: 52.2–59.1) in AFO group ($P = 0.61$). The 3-year overall survival (OS) rates were 75.5% and 58.82%, and 3-year progression-free survival (PFS) rates were 67.5% and 53.92% in non-airflow obstruction and airflow obstruction groups, respectively. The airflow obstruction patients' OS and PFS rates were significantly worse than those of non-obstruction patients ($P < 0.001$ and $P = 0.002$, respectively, Figure 2). Table 3 presents a multivariate Cox regression analysis performed on factors showing significance in the univariate analysis (age, gender, smoking status, surgical approach, pT, pN, G, PNI, LVSI, and anastomotic leakages). Airflow obstruction turned out to be an independent risk factor for OS (Hazard Ratio = 1.66; 95%CI: 1.17–2.35, $P = 0.004$) and PFS (Hazard Ratio = 1.51; 95% CI: 1.1–2.08; $P = 0.01$) in esophageal cancer patients.

## Subgroup survival analysis

Overall survival stratified by several covariates was analyzed. When patients were male ($P = 0.003$), with BMI < 23 kg/m$^2$ ($P < 0.001$), with late-stage cancer (stage III-IVA) ($P = 0.002$), or undergoing open esophagectomy ($P < 0.001$), the overall survival was significantly shorter in AFO group compared with non-AFO group. Other covariates showed no differences in survival between the two groups (Figure 3).

Notably, the 3-year survival rate of airflow obstruction with open surgical procedure or stage III-IVA was 44% and 31%, respectively, which were much lower than those in any other subgroups analyzed.

## Impact of airflow obstruction with BMI < 23 kg/m$^2$ on survival of esophageal cancer patients

Among all baseline variables, BMI was significantly lower in the obstruction group than in non-airflow obstruction group (22.2 vs. 22.9, $P = 0.019$). We evaluated the comprehensive impact of airflow obstruction and BMI on survival. A BMI of 23 kg/m$^2$ is used to distinguish whether a patient is overweight. Patients with both BMI < 23 kg/m$^2$ and airflow

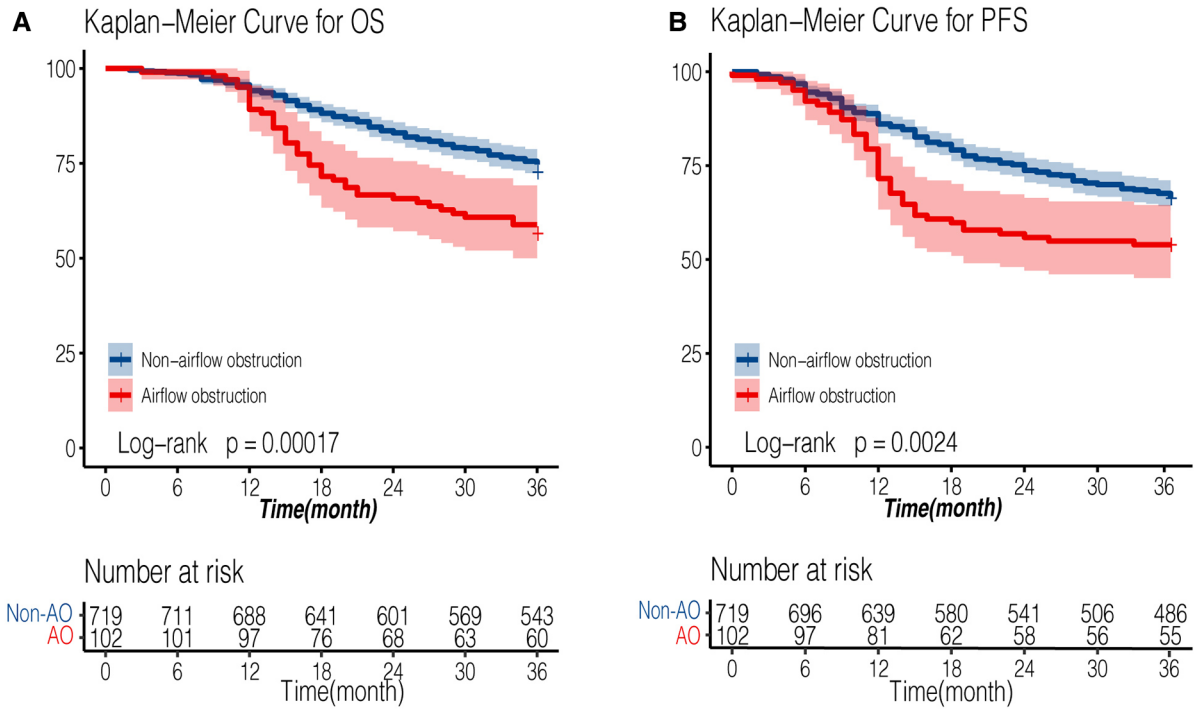


FIGURE 2
Kaplan-Meier survival curve of AFO and non-AFO groups in patients undergoing esophagectomy for esophageal cancers. (A): overall survival curve; (B): progression-free survival curve). AFO: airflow obstruction.

obstruction showed inferior outcomes (3-year OS: 48%, Figure 3), which was significantly worse than that of patients in the other three groups (all $P < 0.05$, Figure 4). However, the BMI value was not related to the overall survival of the entire study population (Table 3).

To further validate this finding, we performed a univariate Cox regression analysis in AFO group (Supplementary Table S2). Variables with $P < 0.05$ in the univariate Cox regression analysis were included in the multivariate Cox proportional splines model to reflect the non-linear relation between all-cause mortality and BMI as a continuous variable. Hazard ratios of mortality decreased more as BMI increased (Supplementary Figure S1B) in airflow obstruction patients, compared with that in the whole population (Supplementary Figure S1A).

## Airflow obstruction promotes lung metastasis in esophageal cancer patients

It was noteworthy that lung metastasis was associated with airflow obstruction (Table 2, $P = 0.01$). The evaluation of risk factors for lung metastasis in esophageal cancer patients is shown in Table 4. In multivariate Cox regression analysis, airflow obstruction was associated with a significantly increased probability (Hazard Ratio = 2.22; 95% CI: 1.31–3.78; $P = 0.003$) of lung metastasis from the primary tumor. The risk for lung metastasis also significantly increased when the pathological N factor was larger than 0 (Hazard Ration = 1.73; 95% CI: 1.07–2.78; $P = 0.024$).

# Discussion

This is a single-center-based retrospective cohort study of patients with esophageal cancer. And it is the first study on the prognosis impact of preoperative airflow obstruction defined as $FEV_1/FVC < LLN$ for esophageal cancer. Our findings suggest that (i) airflow obstruction was observed in 12.4% of patients receiving esophageal cancer surgery, (ii) preoperative airflow obstruction was an independent prognostic factor for 3-year OS and PFS following trans-thoracic esophagectomy. (iii) preoperative airflow obstruction was an independent risk factor for pulmonary metastasis in esophageal cancer.

The impact of airway obstruction on patients' survival outcomes should not be surprising. Trans-thoracic esophagectomy affects the activity of the chest wall and the lung. Meanwhile, the stomach moves upward and squeezes into the lungs after esophagogastrostomy, resulting in limited

TABLE 3 Cox proportional hazards regression models for predictors of overall survival (OS) and progression-free survival (PFS).

| Characteristics | No. | *Univariate analysis* | | *Multivariate analysis* | |
|---|---|---|---|---|---|
| | | HR (95% CI) | *p* value | HR (95% CI) | *p* value |
| ***Overall survival*** | | | | | |
| Airflow Obstruction | 102 | 1.89 (1.35–2.64) | <0.001* | 1.66 (1.17–2.35) | 0.004* |
| Age >70 (vs. ≤70) | 83 | 1.5 (1.03–2.2) | 0.035* | 1.27 (0.86–1.88) | 0.23 |
| Male | 626 | 2.33 (1.59–3.45) | <0.001* | 1.72 (1.12–2.63) | 0.013* |
| BMI ≥23 (vs. <23) | 402 | 1.12 (0.69–1.16) | 0.39 | | |
| Smoker | 355 | 1.31 (1.01–1.7) | 0.046* | 1.06 (0.8–1.41) | 0.69 |
| MIE approach (vs. Open approach) | 389 | 0.62 (0.47–0.81) | <0.001* | 0.69 (0.53–0.92) | 0.011* |
| Complications | 337 | 1.14 (0.88–1.49) | 0.32 | | |
| Anastomotic leakage | 133 | 1.47 (1.07–2.03) | 0.019* | 1.34 (0.96–1.86) | 0.09 |
| Neoadjuvant chemotherapy | 86 | 1.22 (0.81–1.83) | 0.35 | | |
| G3 (vs. G1-2) | 280 | 1.78 (1.37–2.32) | <0.001* | 1.25 (0.95–1.65) | 0.11 |
| PNI | 129 | 2.04 (1.51–2.77) | <0.001* | 1.1 (0.78–1.54) | 0.59 |
| LVSI | 99 | 2.39 (1.74–3.3) | <0.001* | 1.62 (1.14–2.29) | 0.007* |
| pT | | | | | |
| T0-1 | 278 | REF | REF | REF | REF |
| T2 | 206 | 2.22 (1.45–3.41) | <0.001* | 1.5 (0.96–2.35) | 0.077 |
| T3-4 | 337 | 3.87 (2.67–5.61) | <0.001* | 2.21 (1.47–3.29) | <0.001* |
| pN | | | | | |
| N0 | 543 | REF | | REF | |
| N1 | 174 | 2.32 (1.69–3.19) | <0.001* | 1.77 (1.27–2.45) | <0.001* |
| N2-3 | 104 | 4.62 (3.36–6.34) | <0.001* | 2.64 (1.86–3.76) | <0.001* |
| ***Progression-free survival*** | | | | | |
| Airflow Obstruction | 102 | 1.62 (1.18–2.21) | 0.003* | 1.51 (1.1–2.08) | 0.011* |
| Age >70 (vs. ≤70) | 83 | 1.34 (0.94–1.9) | 0.102 | | |
| Male | 626 | 1.78 (1.32–2.44) | <0.001* | 1.39 (1.01–1.92) | 0.043* |
| Smoker | 355 | 1.23 (0.98–1.55) | 0.076 | | |
| MIE approach (vs. Open approach) | 389 | 0.66 (0.51–0.83) | 0.001* | 0.77 (0.60–0.98) | 0.03* |
| Complications | 337 | 1.14 (0.91–1.44) | 0.26 | | |
| Anastomotic leakage | 133 | 1.35 (1.01–1.8) | 0.045* | 1.23 (0.91–1.66) | 0.17 |
| Neoadjuvant chemotherapy | 86 | 1.19 (0.83–1.71) | 0.36 | | |
| G3 (vs. G1-2) | 280 | 1.61 (1.27–2.03) | <0.001* | 1.11 (0.87–1.42) | 0.41 |
| PNI | 129 | 1.95 (1.48–2.56) | <0.001* | 1.05 (0.78–1.42) | 0.74 |
| LVSI | 99 | 1.95 (1.8–3.19) | <0.001* | 1.563 (1.2–2.22) | 0.002* |
| pT | | | | | |
| T0-1 | 278 | REF | | REF | |
| T2 | 206 | 1.96 (1.36–2.83) | <0.001* | 1.48 (1.02–2.17) | 0.041* |
| T3-4 | 337 | 3.74 (2.73–5.12) | <0.001* | 2.43 (1.72–3.42) | <0.001* |
| pN | | | | | |
| N0 | 543 | REF | | REF | |
| N1 | 174 | 2.18 (1.65–2.87) | <0.001* | 1.67 (1.25–2.22) | <0.001* |
| N2-3 | 104 | 4.05 (3.05–5.4) | <0.001* | 2.44 (1.78–3.35) | <0.001* |

HR, hazard ratio; CI, confidence interval; MIE, minimally invasive esophagectomy; pT, pathological T factor; pN, pathological N factor; PNI, perineural invasion; LVSI, lymph-vascular space invasion.
*$P < 0.05$.

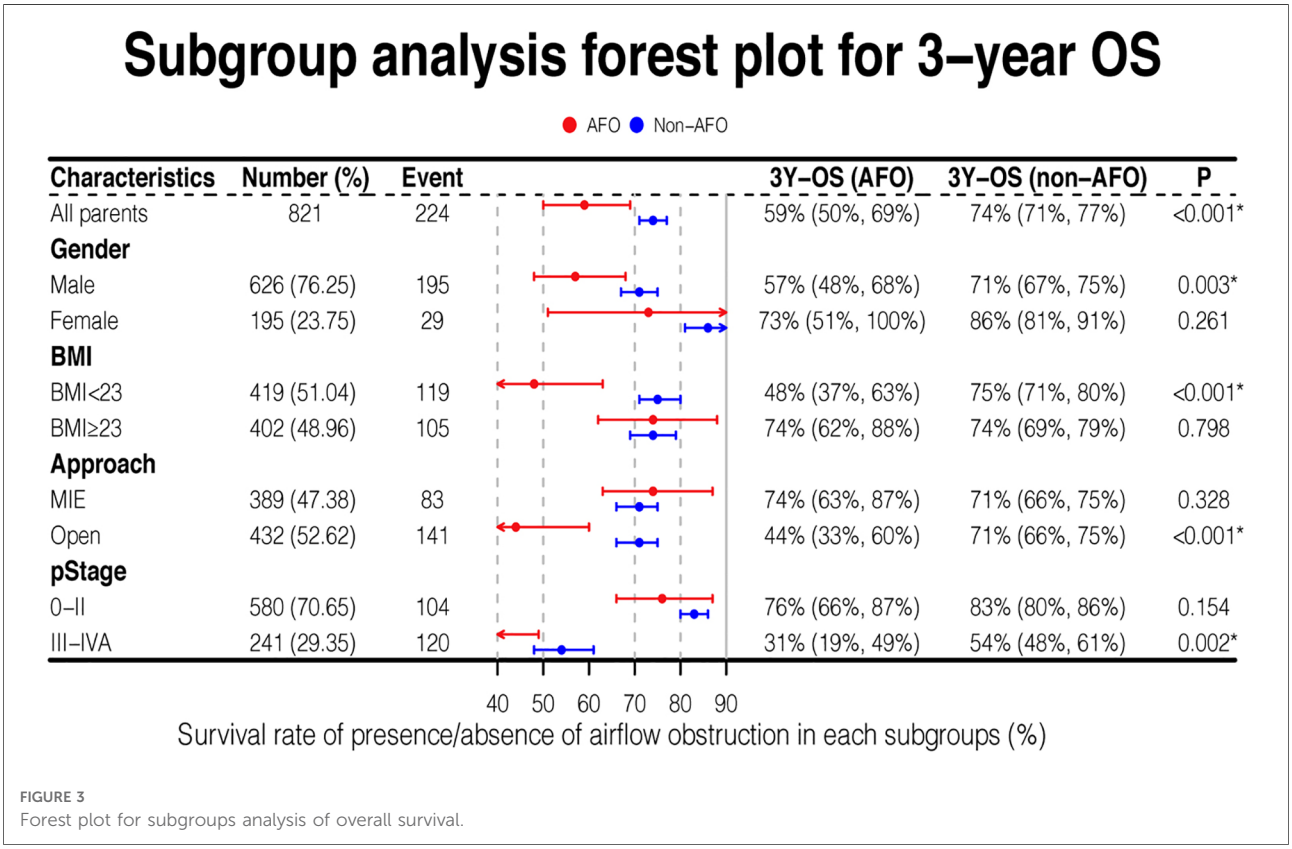


FIGURE 3
Forest plot for subgroups analysis of overall survival.

pulmonary dilatation and accelerated lung function decline. Patients with chronic airflow obstruction diseases (COPD and asthma, for instance) may be more susceptible to anastomotic leakages and infections, which detrimentally affect survival by delaying recovery or leading to death (21, 22).

Subgroup analysis shed light on the most sensitive population to airflow obstruction. Minimally invasive esophagectomy (MIE) could reduce the response of the organism, accelerate recovery and maintain postoperative pulmonary function (23, 24). Patients with airflow obstruction may particularly benefit from MIE. Moreover, airflow obstruction worsened survival of stage III-IVA esophageal cancer; but showed no difference in patients with stage 0-II cancer. This is probably because late-stage cancer patients have deteriorating disease manifestations and declining quality of life (25, 26). The presence of airflow obstruction worsens the cognitive and overall status at certain levels (27, 28), playing an adjunctive role in the lethal effects of EC. But in the early stages, the follow-up was relatively short, and most of them did not experience the outcome event. The sex difference might be because insufficient female patients led to investigation bias.

A previous study demonstrated that patients with lower BMI had a faster $FEV_1$/FVC decline and more symptoms than patients with higher BMI (29). In line with these prior results (30, 31), patients with airflow obstruction in our study had lower BMI. It is noteworthy that patients with airflow obstruction but BMI $\geq$23 kg/m$^2$ exhibited as good survival outcomes as the non-airflow obstruction group, which suggested higher BMI could be protective in esophageal cancer patients complicated with airflow obstruction. Therefore, we assume that BMI or overall nutrition status could partly explain our findings on survival outcomes.

Another interesting phenomenon was that airway obstruction facilitated the lung spread of esophageal cancer. This finding echoes the impact of smoking (32) since smoking is highly correlated to airflow obstruction. The "seed-and-soil hypothesis" partially explains this finding (33, 34). Airway obstruction usually coexists with the remodeling of the airway epithelium and alterations of the distribution of inflammatory cells, providing an ideal micro-environment (soil) for tumor cells (seed) colonization and growth (35). Therefore, our findings shed new light on the mechanism of lung metastasis of esophageal cancer.

Unfortunately, in our study, only 25 (25/102, 24.5%) were diagnosed with chronic obstructive airway diseases before the esophagectomy. Almost all patients (95/102, 93.1%) were without sustained lung-directed therapy. Although

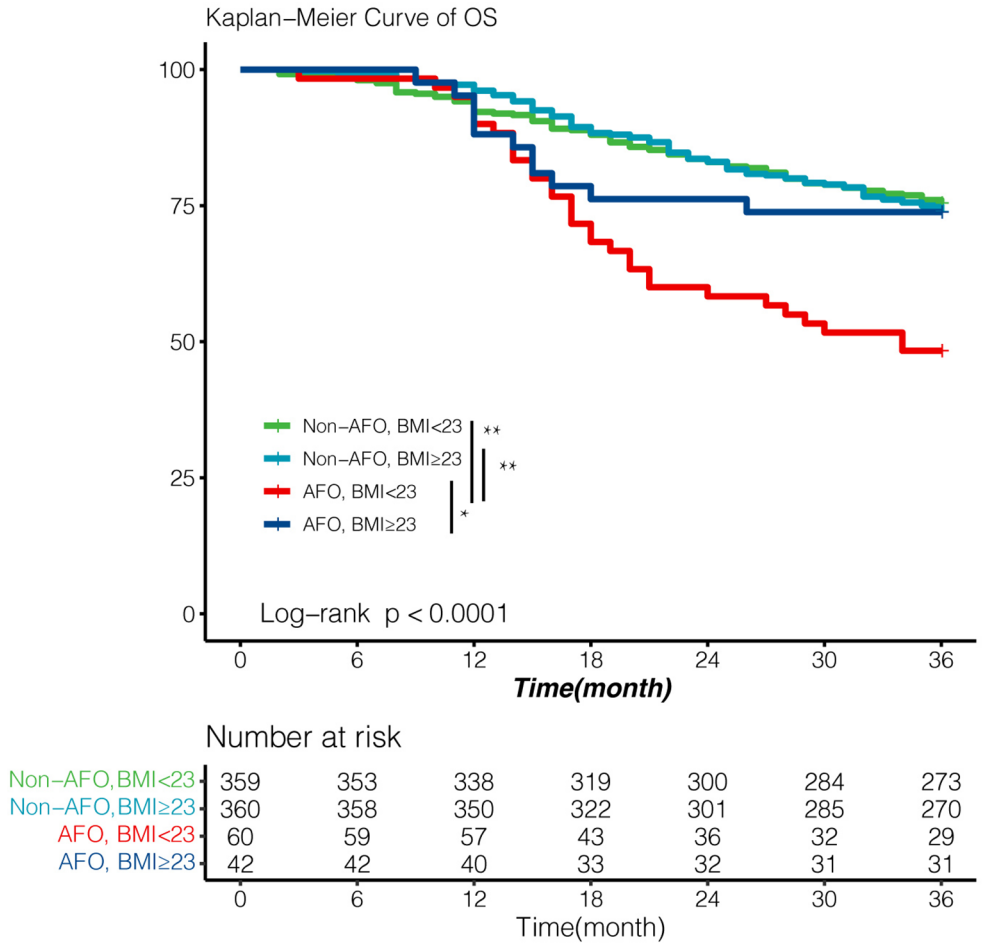


FIGURE 4
Survival according to airflow obstruction and BMI with cut-off value 23 kg/m$^2$. Patients with both airflow obstruction and BMI < 23 showed significantly poor OS (3-year OS; 48%). Survival outcomes of other three patients were essentially equivalent (3-year OS; 75%, 74.0%, 74% respectively). Survival was analyzed by Kaplan–Meier method using the log-rank test. AFO: airflow obstruction. BMI:body mass index. *$p$ value < 0.05, **$p$ value < 0.01.

**TABLE 4** Univariate and multivariate Cox proportional hazards regression analysis for the evaluation of risk factors for lung metastasis within 3 years after esophagectomy.

| | Events | Univariate HR (95% CI) | *P* value | Multivariate HR (95% CI) | *P* value |
|---|---|---|---|---|---|
| Airflow Obstruction | 18 | 2.40 (1.4–4) | <0.001* | 2.22 (1.31–3.78) | 0.005* |
| Male | 60 | 1.2 (0.67–2.08) | 0.52 | | |
| MIE approach (vs. Open approach) | 31 | 0.77 (0.48–1.19) | 0.23 | | |
| G3 (vs. G1-2) | 29 | 1.21 (0.75–1.9) | 0.45 | | |
| pT3-4 (vs. pT0-2) | 41 | 1.7 (1.1–2.7) | 0.02* | 1.4 (0.86–2.3) | 0.17 |
| pN1-3 (vs. pN0) | 38 | 2 (1.3–3.2) | <0.001* | 1.73 (1.07–2.78) | 0.024* |
| PNI | 17 | 1.61 (0.91–2.77) | 0.09 | 1.19 (0.67–2.11) | 0.54 |
| LVSI | 12 | 1.39 (0.77–2.56) | 0.29 | | |
| Anastomotic leakage | 16 | 1.4 (0.8–2.4) | 0.25 | | |
| Pulmonary complications | 17 | 1.4 (0.82–2.4) | 0.22 | | |

HR, hazard ratio; CI, confidence interval; MIE, minimally invasive esophagectomy; pT, pathological T factor; pN, pathological N factor; PNI, perineural invasion; LVSI, lymph-vascular space invasion.
*$P$ < 0.05.

undiagnosed airflow obstruction subjects appeared healthier than those with a diagnosis, their prognosis was worse than subjects without airflow obstruction[15]. Our work suggests that preoperative airflow obstruction and potential obstructive airway diseases should be given more attention. Perioperative and long-term airway intervention deserves further investigation to improve survival outcomes.

There are some limitations to our study. First, the median follow-up duration was 54 months in the whole study population, while more extended follow-up periods may provide detailed information on EC prognosis, especially in stage 0-II patients. Secondly, the sample size of patients receiving neoadjuvant therapy was not enough. The interaction between airflow obstruction and neoadjuvant treatment remains to be demonstrated. Finally, 93.5% of patients in our cohort were with esophageal squamous cell carcinoma, whose BMI was generally lower than average (36). It remains unclear whether our conclusions apply to western countries, where adenocarcinoma is the primary pathological type.

## Conclusion

Airflow obstruction is a common comorbidity in patients with esophageal cancer. Patients with airflow obstruction had more postoperative complications and shorter 3-year OS and PFS after trans-thoracic surgery for esophageal cancer. BMI or overall nutrition status could partly explain these effects. More attention is needed to manage airflow obstruction in esophageal cancer patients comprehensively. We should incorporate the patient's respiratory condition into the surgical decision-making process to reach a better prognosis.

## Data availability statement

The raw data supporting the conclusions of this article will be made available by the authors, without undue reservation.

## Ethics statement

The studies involving human participants were reviewed and approved by The Ethics Committee of Zhongshan Hospital, Fudan University. The patients/participants provided their written informed consent to participate in this study.

## Author contributions

KL: data collection, data analysis, and manuscript writing; XW: data collection, data management. TW: manuscript editing and data management. ZG & YS: data collection; DY & HW: project development, data management, and manuscript editing. All authors contributed to the article and approved the submitted version.

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## Conflict of interest

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

## Publisher's note



## Supplementary material

The Supplementary Material for this article can be found online at: https://www.frontiersin.org/articles/10.3389/fsurg.2022.966340/full#supplementary-material.

# References


1. Lagergren J, Smyth E, Cunningham D, Lagergren P. Oesophageal cancer. *Lancet*. (2017) 390:2383–96. doi: 10.1016/S0140-6736(17)31462-9

2. Huang J, Koulaouzidis A, Marlicz W, Lok V, Chu C, Ngai CH, et al. Global burden, risk factors, and trends of esophageal cancer: an analysis of cancer registries from 48 countries. *Cancers (Basel)*. (2021) 13:141. doi: 10.3390/cancers13010141

3. Klevebro F, Elliott JA, Slaman A, Vermeulen BD, Kamiya S, Rosman C, et al. Cardiorespiratory comorbidity and postoperative complications following esophagectomy: a European multicenter cohort study. *Ann Surg Oncol*. (2019) 26:2864–73. doi: 10.1245/s10434-019-07478-6

4. Weksler B, Sullivan JL. Survival after esophagectomy: a propensity-matched study of different surgical approaches. *Ann Thorac Surg*. (2017) 104:1138–46. doi: 10.1016/j.athoracsur.2017.04.065

5. Rice TW, Patil DT, Blackstone EH. 8th Edition AJCC/UICC staging of cancers of the esophagus and esophagogastric junction: application to clinical practice. *Ann Cardiothorac Surg*. (2017) 6:119–30. doi: 10.21037/acs.2017.03.14

6. Shah MA, Kennedy EB, Catenacci DV, Deighton DC, Goodman KA, Malhotra NK, et al. Treatment of locally advanced esophageal carcinoma: aSCO guideline. *J Clin Oncol*. (2020) 38:2677–94. doi: 10.1200/JCO.20.00866

7. Yoshida N, Harada K, Iwatsuki M, Baba Y, Baba H. Precautions for avoiding pulmonary morbidity after esophagectomy. *Ann Gastroenterol Surg*. (2020) 4:480–4. doi: 10.1002/ags3.12354

8. Ho CH, Chen YC, Wang JJ, Liao KM. Incidence and relative risk for developing cancer among patients with COPD: a nationwide cohort study in Taiwan. *BMJ Open*. (2017) 7:e013195. doi: 10.1136/bmjopen-2016-013195

9. Molena D, Mungo B, Stem M, Lidor AO. Incidence and risk factors for respiratory complications in patients undergoing esophagectomy for malignancy:a NSQIP analysis. *Semin Thorac Cardiovasc Surg*. (2014) 26:287–94. doi: 10.1053/j.semtcvs.2014.12.002

10. Wei R, Dong W, Shen H, Ni Y, Zhang T, Wang Y, et al. Predictive effects of lung function test on postoperative pneumonia in squamous esophageal cancer. *Sci Rep*. (2016) 6:23636. doi: 10.1038/srep23636

11. Ohi M, Toiyama Y, Omura Y, Ichikawa T, Yasuda H, Okugawa Y, et al. Risk factors and measures of pulmonary complications after thoracoscopic esophagectomy for esophageal cancer. *Surg Today*. (2019) 49:176–86. doi: 10.1007/s00595-018-1721-0

12. Sugawara K, Mori K, Okumura Y, Yagi K, Aikou S, Uemura Y, et al. Preoperative low vital capacity influences survival after esophagectomy for patients with esophageal carcinoma. *World J Surg*. (2020) 44:2305–13. doi: 10.1007/s00268-020-05450-0

13. Pellegrino R, Viegi G, Brusasco V, Crapo RO, Burgos F, Casaburi R, et al. Interpretative strategies for lung function tests. *Eur Respir J*. (2005) 26:948–68. doi: 10.1183/09031936.05.00035205

14. Halpin DMG, Criner GJ, Papi A, Singh D, Anzueto A, Martinez FJ, et al. Global initiative for the diagnosis, management, and prevention of chronic obstructive lung disease. The 2020 GOLD science committee report on COVID-19 and chronic obstructive pulmonary disease. *Am J Respir Crit Care Med*. (2021) 203:24–36. doi: 10.1164/rccm.202009-3533SO

15. Quanjer PH, Enright PL, Miller MR, Stocks J, Ruppel G, Swanney MP, et al. The need to change the method for defining mild airway obstruction. *Eur Respir J*. (2011) 37:720–2. doi: 10.1183/09031936.00135110

16. Jian W, Gao Y, Hao C, Wang N, Ai T, Liu C, et al. Reference values for spirometry in Chinese aged 4-80 years. *J Thorac Dis*. (2017) 9:4538–49. doi: 10.21037/jtd.2017.10.110

17. Low DE, Alderson D, Cecconello I, Chang AC, Darling GE, D'Journo XB, et al. International consensus on standardization of data collection for complications associated with esophagectomy: esophagectomy complications consensus group (ECCG). *Ann Surg*. (2015) 262:286–94. doi: 10.1097/SLA.0000000000001098

18. Schemper M, Smith TL. A note on quantifying follow-up in studies of failure time. *Control Clin Trials*. (1996) 17:343–6. doi: 10.1016/0197-2456(96)00075-X

19. Durrleman S, Simon R. Flexible regression models with cubic splines. *Stat Med*. (1989) 8:551–61. doi: 10.1002/sim.4780080504

20. Govindarajulu US, Spiegelman D, Thurston SW, Ganguli B, Eisen EA. Comparing smoothing techniques in cox models for exposure-response relationships. *Stat Med*. (2007) 26:3735–52. doi: 10.1002/sim.2848

21. Andreou A, Biebl M, Dadras M, Struecker B, Sauer IM, Thuss-Patience PC, et al. Anastomotic leak predicts diminished long-term survival after resection for gastric and esophageal cancer. *Surgery*. (2016) 160:191–203. doi: 10.1016/j.surg.2016.02.020

22. Saunders JH, Yanni F, Dorrington MS, Bowman CR, Vohra RS, Parsons SL, et al. Impact of postoperative complications on disease recurrence and long-term survival following oesophagogastric cancer resection. *Br J Surg*. (2020) 107:103–12. doi: 10.1002/bjs.11318

23. Otani T, Ichikawa H, Hanyu T, Ishikawa T, Kano Y, Kanda T, et al. Long-Term trends in respiratory function after esophagectomy for esophageal cancer. *J Surg Res*. (2020) 245:168–78. doi: 10.1016/j.jss.2019.07.040

24. Briez N, Piessen G, Torres F, Lebuffe G, Triboulet JP, Mariette C. Effects of hybrid minimally invasive oesophagectomy on major postoperative pulmonary complications. *Br J Surg*. (2012) 99:1547–53. doi: 10.1002/bjs.8931

25. Bracken-Clarke D, Farooq AR, Horgan AM. Management of locally advanced and metastatic esophageal cancer in the older population. *Curr Oncol Rep*. (2018) 20:99. doi: 10.1007/s11912-018-0745-3

26. Lagergren P, Avery KN, Hughes R, Barham CP, Alderson D, Falk SJ, et al. Health-related quality of life among patients cured by surgery for esophageal cancer. *Cancer* (2007) 110:686–93. doi: 10.1002/cncr.22833

27. Odeyemi YE, Meda E, Ogundipe F, Russ E, Mehari A, Obisesan T, et al. Airflow obstruction, cognitive function and mortality in a US national cohort: nHANES-III. *Clin Respir J*. (2018) 12:1141–9. doi: 10.1111/crj.12643

28. Burney PG, Hooper R. Forced vital capacity, airway obstruction and survival in a general population sample from the United States. *Thorax*. (2011) 66:49–54. doi: 10.1136/thx.2010.147041

29. Luoto J, Pihlsgård M, Wollmer P, Elmståhl S. Relative and absolute lung function change in a general population aged 60–102 years. *Eur Respir J*. (2019) 53:1701812. doi: 10.1183/13993003.01812-2017

30. Burney P, Patel J, Minelli C, Gnatiuc L, Amaral AFS, Kocabaş A, et al. Prevalence and population attributable risk for chronic airflow obstruction in a large multinational study. *Am J Respir Crit Care Med*. (2020) 203:1353–65. doi: 10.1164/rccm.202005-1990OC

31. Graff S, Bricmont N, Moermans C, Henket M, Paulus V, Guissard F, et al. Clinical and biological factors associated with irreversible airway obstruction in adult asthma. *Respir Med*. (2020) 175:106202. doi: 10.1016/j.rmed.2020.106202

32. Abrams JA, Lee PC, Port JL, Altorki NK, Neugut AI. Cigarette smoking and risk of lung metastasis from esophageal cancer. *Cancer Epidemiol Biomarkers Prev*. (2008) 17:2707–13. doi: 10.1158/1055-9965.EPI-08-0232

33. Paget S. The distribution of secondary growths in cancer of the breast. *Cancer Metastasis Rev*. (1989) 8:98–101. doi: 10.1016/s0140-6736(00)49915-0

34. Fidler IJ. The pathogenesis of cancer metastasis: the "seed and soil" hypothesis revisited. *Nat Rev Cancer*. (2003) 3:453–8. doi: 10.1038/nrc1098

35. Houghton AM. Mechanistic links between COPD and lung cancer. *Nat Rev Cancer*. (2013) 13:233–45. doi: 10.1038/nrc3477

36. Renehan AG, Tyson M, Egger M, Heller RF, Zwahlen M. Body-mass index and incidence of cancer: a systematic review and meta-analysis of prospective observational studies. *Lancet*. (2008) 371:569–78. doi: 10.1016/S0140-6736(08)60269-X

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# Review of the status of neoadjuvant therapy in HER2-positive breast cancer

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**Purpose:** The development of human epidermal growth factor receptor 2 (HER2)-directed therapies has revolutionized the treatment of HER2-positive breast cancer. The aim of this article is to review the continually evolving treatment strategies in the neoadjuvant setting of HER2-positive breast cancer, as well as the current challenges and future perspectives.

**Methods:** Searches were undertaken on PubMed and Clinicaltrials.gov for relevant publications and trials.

**Findings:** The current standard of care in high-risk HER2-positive breast cancer is to combine chemotherapy with dual anti-HER2 therapy, for a synergistic anti-tumor effect. We discuss the pivotal trials which led to the adoption of this approach, as well as the benefit of these neoadjuvant strategies for guiding appropriate adjuvant therapy. De-escalation strategies are currently being investigated to avoid over treatment, and aim to safely reduce chemotherapy, while optimizing HER2-targeted therapies. The development and validation of a reliable biomarker is essential to enable these de-escalation strategies and personalization of treatment. In addition, promising novel therapies are currently being explored to further improve outcomes in HER2-positive breast cancer.

KEYWORDS

**neoadjuvant therapy, breast cancer, HER2 (human epidermal growth factor 2), targeted therapy, biomarker, antibody-drug-conjugates**

## Introduction

Human epidermal growth factor receptor 2 (HER2) is overexpressed and/or amplified in 15-20% of all breast cancers. Before the advent of HER2-directed therapies, this subtype was associated with an aggressive clinical course and poor outcomes (1, 2). The introduction of trastuzumab, the first humanized anti-HER2 monoclonal antibody, transformed the treatment of HER2-positive breast cancer. The benefit of neoadjuvant breast cancer treatment with chemotherapy, endocrine therapy and/or targeted therapy is well

established to downstage disease, improve resectability and potentially reduce the extent of breast and axillary surgery (3–5). Specifically, in the HER2-positive subtype, additional benefits of neoadjuvant systemic therapy have been appreciated. These include the potential to individualize adjuvant therapy options based on pathological response and to provide information about tumor status *in vivo*, allowing for escalation or de-escalation of therapy, as guided by response biomarkers. Thus, the current standard of care in patients with high-risk HER2-positive breast cancer is a combination of chemotherapy combined with dual anti-HER2 therapy (6, 7). This review will discuss the evolving standard of care in the neoadjuvant setting of HER2-positive breast cancer, as well as the challenges and future perspectives.

## Review of neoadjuvant trials

Neoadjuvant therapy is the current standard of care for treating ≥T2 or node-positive HER2-positive breast cancer. Pathological complete response (pCR) is most commonly defined as the absence of residual invasive cancer of the complete resected breast specimen and all sampled regional lymph nodes following completion of neoadjuvant systemic therapy (ypT0/Tis ypN0) (8). pCR at surgery is correlated with favorable patient outcomes, particularly in HER2-positive, hormone-receptor (HR)-negative breast cancer, as demonstrated by the CTNeoBC pooled analysis. This meta-analysis, performed by the FDA, included 11,955 patients across 12 neoadjuvant trials, with a minimum follow-up of 3 years, to evaluate pCR as a surrogate endpoint for improved long-term outcomes in breast cancer. Across all subgroups, pCR was associated with improved event-free survival (EFS) (HR 0.48; 95% CI 0.43-0.54) and overall survival (OS) (HR 0.36; 95% CI 0.31-0.42). Three trials were included for HER2-positive breast cancer: NOAH, TECHNO and GeparQuattro (9). Several additional meta-analyses have since supported the value of pCR as an informative surrogate biomarker for enhanced survival in HER2-positive breast cancer (10–12).

Trastuzumab is a monoclonal antibody against HER2 that binds to an extracellular domain of this receptor and prevents ligand-independent HER2-mediated signaling (13). Following its success in treating advanced and early-stage HER2-positive disease, multiple neoadjuvant trials that combine chemotherapy with trastuzumab have been performed. The NOAH trial, for example, reported that the addition of trastuzumab to neoadjuvant chemotherapy had a significant improvement on both pCR and EFS when compared to chemotherapy alone (14). Similar improvements in pCR were seen with the addition of trastuzumab to a chemotherapy backbone in the TECHNO trial and GeparQuattro study (15, 16).

Pertuzumab binds to the extracellular domain II of HER2, which results in ligand-dependent HER2–HER3 dimerization (17). This mechanism of action is complementary to that of trastuzumab. In the NeoSphere trail, a pCR rate of approximately 45% was observed in patients treated with pertuzumab plus trastuzumab and docetaxel, compared to those who received only trastuzumab and docetaxel (29%) (35). This combination of pertuzumab with trastuzumab and docetaxel was also investigated in the CLEOPATRA trial, which reported a significant overall survival benefit (56.5 months vs 40.8 months) (18). Following these trials, dual HER2-blockade with trastuzumab and pertuzumab in combination with standard neoadjuvant chemotherapy became the standard of care (7, 19).

Lapatinib is a dual reversible tyrosine kinase inhibitor that selectively targets and inhibits HER2 and epidermal growth factor receptor (EGFR) (20). Lapatinib has demonstrated activity in HER2-positive metastatic breast cancer that had progressed on trastuzumab-containing therapy (21). In the Cher-LOB trial, patients treated with lapatinib and trastuzumab plus chemotherapy showed a relative 80% increase in pCR rate, compared to treatment with either trastuzumab or lapatinib plus chemotherapy (22). Additionally, the CALGB 40601 trial showed improved 7-year relapse-free survival and OS (23). Despite several studies showing improved pCR rates with the addition of lapatinib to trastuzumab and chemotherapy in the neoadjuvant setting, these long-term outcomes have not been consistent across trials (24–27). The inconsistency of long-term outcomes, along with the less favorable adverse event profile associated with the addition of lapatinib, has prevented it from becoming a currently recommended neoadjuvant treatment.

Both anthracycline and non-anthracycline-based chemotherapy regimens are well established as neoadjuvant treatments of HER2-positive breast cancer. Combination treatment with anthracyclines and trastuzumab can have significant side effects in patients, including febrile neutropenia and cardiotoxicity. Multiple trials have explored the feasibility of treating these patients with anthracycline-free regimes. The TRAIN-2 trial reported high pCR rates after neoadjuvant chemotherapy with or without anthracyclines plus dual-HER2 blockade. No significant difference was seen in either pCR or patient outcomes between the two groups (28). In addition, the TRYPHAENA trial showed similar efficacy for anthracycline-free compared to anthracycline-containing regimens together with standard anti-HER2 therapy. Cardiac safety was the primary endpoint: left ventricular systolic dysfunction (LVSD) incidence was low (5.6%) in the neoadjuvant setting in the anthracycline-containing arm (29). Furthermore, the BERENICE trial demonstrated cardiac safety in both dose-dense and standard anthracycline-containing regimens in combination with trastuzumab and pertuzumab (30, 31).

Given the success of neoadjuvant systemic chemotherapy with dual HER2-blockade, achieving pCR rates of up to 65% in some studies (28, 29), the possibility of replacing chemotherapy with an agent associated with less toxicity was explored. The phase III KRISTINE study compared neoadjuvant trastuzumab emtansine (T-DM1), an antibody-drug conjugate, plus pertuzumab, with conventional systemic chemotherapy plus dual HER2-blockade. The results showed that the proportion of patients who achieved pCR was significantly greater in patients receiving traditional neoadjuvant chemotherapy plus trastuzumab and pertuzumab than those who received T-DM1 plus pertuzumab (56% vs 44%) (32). The results of these neoadjuvant clinical trials are summarised in Table 1.

To prevent the potential of over-treatment in patients with low-risk HER2-positive breast cancer, the APT trial was designed. This study included patients with ≤3cm, node-negative, HER2-positive tumors. This trial showed excellent outcomes with adjuvant paclitaxel for 12 weeks plus 12 months of trastuzumab, with a 3-year IDFS of 98.7% and 7-year IDFS of 93% (41). Thus, primary surgery combined with adjuvant therapy should be offered to these patients, providing an effective de-escalated treatment regime.

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TABLE 1 Neoadjuvant Trials in HER2-positive breast cancer.

| Trial | Phase | Treatment | Treatment arms | Survival | N | pCR rate |
|---|---|---|---|---|---|---|
| Trastuzumab | | | | | | |
| NOAH (14) | III | Trastuzumab | DTCMF + H | EFS (3y) 71%<br>OS (3y) 87% | 117 | 38% |
| | | | DTCMF | EFS (3y) 56%<br>OS (3y) 79% | 118 | 19% |
| GeparQuattro (15) | III | Trastuzumab | H + EC → H + Dc | | 146 | 33% |
| | | | H + EC → H + DcCp | | 47 | 31% |
| | | | H + EC → H + Dc → H + Cp | | 136 | 35% |
| TECHNO (16) | II | Trastuzumab | EC → HT | DFS (3y) 77.9%<br>OS (3y) 89.4% | 217 | 39% |
| Trastuzumab + Lapatinib | | | | | | |
| NeoALTTO (24) | III | Lapatinib + Trastuzumab | LH → HT | EFS (6y) 74%<br>OS (6y) 85% | 68 | 47% |
| | | | H → HT | EFS (6y) 67%<br>OS (6y) 82% | 40 | 28% |
| | | | L → HT | EFS (6y) 67%<br>OS (6y) 79% | 30 | 20% |
| CHER-LOB (22) | II | Lapatinib + Trastuzumab | LH → TFEC + LH | RFS (5y) 86% | 46 | 47% |
| | | | H → TFEC + H | RFS (5y) 78% | 36 | 25% |
| | | | L → TFEC + L | RFS (5y) 77% | 38 | 26% |
| CALGB 40601 (23) | III | Lapatinib + Trastuzumab | THL | RFS (7y) 93%<br>OS (7y) 96% | 118 | 57% |
| | | | TH | RFS (7y) 79%<br>OS (7y) 88% | 120 | 45% |
| | | | TL | RFS (7y) 69%<br>OS (7y) 84% | 67 | 30% |
| NSABP B-41 (33) | III | Trastuzumab + Lapatinib | DC + H → T + H | DFS (5y) 84.3%<br>OS (5y) 94.5% | 177 | 49.4% |
| | | | DC + L → T + H | DFS (5y) 78.6%<br>OS (5y) 89.4% | 159 | 47.4% |
| | | | DC + LH → T + H | DFS (5y) 90%<br>OS (5y) 95.7% | 165 | 60.2% |
| Lapatinib | | | | | | |
| GeparQuinto (34) | III | Lapatinib | ECH → Dc + H | DFS (3y) 84.8%<br>OS (3y) 91.7% | 307 | 30.3% |
| | | | ECL → Dc + L | DFS (3y) 83.7%<br>OS (3y) 93.6% | 308 | 22.7% |
| Trastuzumab + Pertuzumab | | | | | | |
| NeoSphere (35) | II | Pertuzumab + Trastuzumab | H + Dc | PFS (5y) 81%<br>DFS (5y) 81% | 107 | 29% |
| | | | HP + Dc | PFS (5y) 86%<br>DFS (5y) 84% | 107 | 46% |
| | | | HP | PFS (5y) 73%<br>DFS (5y) 80% | 107 | 17% |
| | | | P + Dc | PFS (5y) 73%<br>DFS (5y) 75% | 96 | 24% |

*(Continued)*

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TABLE 1 Continued

| Trial | Phase | Treatment | Treatment arms | Survival | N | pCR rate |
|---|---|---|---|---|---|---|
| TRYPHAENA (29) | II | Pertuzumab + Trastuzumab | FEC + HP → Dc + HP | DFS (3y) 87%<br>PFS (3y) 89%<br>OS (3y) 94% | 73 | 62% |
| | | | FEC → Dc + HP | DFS (3y) 88%<br>PFS (3y) 89%<br>OS (3y) 94% | 75 | 57% |
| | | | Dc + Cb + HP | DFS (3y) 90%<br>PFS (3y) 87%<br>OS (3y) 93% | 77 | 66% |
| GeparSepto (36) | III | Pertuzumab + Trastuzumab | T + HP → EC + HP | iDFS (4y) 89% | 199 | 54% |
| | | | Nab-T + HP → EC + HP | | 197 | 62% |
| ADAPT HER2+/HR- (37) | II | Pertuzumab + Trastuzumab | HP | iDFS (5y) 87%<br>OS (5y) 94% | 92 | 34% |
| | | | HP + T | iDFS (5y) 98%<br>OS (5y) 98% | 42 | 90% |
| BERENICE (30, 38) | II | Pertuzumab + Trastuzumab | dd D + C → T + HP | EFS (5y) 91%<br>OS (5y) 96% | 199 | 62% |
| | | | FEC → Dc + HP | EFS (5y) 89%<br>OS (5y) 94% | 201 | 61% |
| TRAIN-2 (28) | III | Pertuzumab + Trastuzumab | FEC (x3) → TCb + HP (x6) | EFS (3y) 93%<br>OS (3y) 98% | 211 | 67% |
| | | | TCb + HP (x9) | EFS (3y) 94%<br>OS (3y) 98% | 206 | 68% |
| PHERGain (39) | II | Pertuzumab + Trastuzumab | DcCb + HP | | 71 | 58% |
| | | | HP | | 285 | 35% |
| PREDIX HER2 (40) | II | Pertuzumab + Trastuzumab | Dc + HP | | 99 | 46% |
| T-DM1 + Pertuzumab | | | | | | |
| KRISTINE (32) | III | T-DM1 + Pertuzumab | T-DM1 + P | EFS (3y) 85%<br>iDFS (3y) 93% | 223 | 44% |
| | | | DcCb + HP | EFS (3y) 94%<br>iDFS (3y) 92% | 221 | 56% |

H- trastuzumab; P- pertuzumab; T- paclitaxel; D- doxorubicin; C- cyclophosphamide; M- methotrexate; F- fluorouracil; L- lapatinib; E- epirubicin; Dc- docetaxel; Cp- capecitabine; Cb- carboplatin, Nab-T- nab-paclitaxel; dd- dose dense; T-DM1- trastuzumab emtansine.

## Adjuvant therapy in the context of neoadjuvant strategy/according to pCR status

Following completion of neoadjuvant therapy, subsequent adjuvant therapies can be guided by pCR status after surgery.

In patients who achieve pCR, current guidelines recommend continuing trastuzumab to complete a total of 12 months of anti-HER2 therapy (6, 42). Patients with initially node-positive disease should also continue pertuzumab for the remainder of the year, based on the findings of the adjuvant APHINITY trial. This trial concluded that the addition of pertuzumab in the adjuvant setting may significantly improve invasive disease-free survival in patients with node-positive disease (43). However, no statistically significant difference was seen in OS after a median follow-up of 8.4 years (44).

In patients who do not achieve pCR, adjuvant therapy with T-DM1 should be offered instead of trastuzumab monotherapy. This recommendation is based on the results from the KATHERINE trial. In this phase III trial, patients with residual invasive tumors after neoadjuvant therapy were randomly assigned to received either adjuvant T-DM1 or trastuzumab for 14 cycles. Treatment with T-DM1 significantly improved invasive disease-free-survival (iDFS) compared to treatment with trastuzumab (88.3% vs. 77.0%, respectively, HR 0.50, 95% CI, 0.39–0.64; $p < 0.001$) (45).

Adjuvant treatment with T-DM1 in stage I HER2-positive breast cancer was investigated in the ATEMPT trial. This trial aimed to establish if adjuvant T-DM1 would be associated with less toxicity than paclitaxel plus trastuzumab without compromising invasive disease-free-survival (iDFS). Although one year of T-DM1 had a 3-year iDFS of 97.8%, T-DM1 failed to demonstrate reduced toxicity compared to paclitaxel and trastuzumab (46).

The KAITLIN study was another trial which aimed to replace taxanes and trastuzumab with T-DM1. In this trial, patients with node-positive or high-risk node-negative (HR negative and tumor size >2cm) HER2-positive breast cancer were randomly assigned to

anthracycline chemotherapy followed by trastuzumab and a taxane plus pertuzumab or anthracycline chemotherapy followed by T-DM1 plus pertuzumab. The results showed no significant difference in 3-year iDFS rate between the two arms of the study (47).

## Duration of anti-HER2 therapy

The current standard of care is to complete 12 months of anti-HER2 therapy. The benefit of this therapy was demonstrated in the crucial HERA, NCCTG N9831, NSABP B-31 and BCIRG-006 trials. It was shown that adjuvant trastuzumab with standard chemotherapy reduced the relative risk of death by up to 30% and the relative risk of recurrence by up to 40% (48–52).

The HERA trial demonstrated that a longer duration of the same anti-HER2 therapy did not improve efficacy, in which two years was compared to one year of trastuzumab treatment. In those who received two years of therapy, no additional benefit in disease free survival (DFS) was seen and associated with a higher rate of cardiotoxicity (48).

Given the effectiveness of anti-HER2 therapy, multiple trials were designed to evaluate the efficacy of reduced duration of treatment.

The PHARE, HORG and PERSEPHONE trials compared 6 months to 12 months of trastuzumab treatment (53–55). PERSEPHONE is the only trial to date to have reached its non-inferiority endpoint. In this trial of 4089 patients, after a median follow-up of 5.4 years, those assigned to 6 months of trastuzumab therapy experienced non-inferior 4-year DFS rates compared to those receiving 12 months (89.4 versus 89.8 percent, respectively; HR 1.07, 95% CI 0.93-1.24), with less cardiotoxicity leading to discontinuation of trastuzumab (55).

A shortened course of 9-weeks of trastuzumab therapy was evaluated in the SOLD and ShortHER trials. These trials failed to reach the non-inferiority endpoint for DFS (56, 57). Despite being unable to claim non-inferiority, recently presented follow-up data of the ShortHER trial confirmed favorable long-term outcomes in terms of OS and DFS with a 9 week course of trastuzumab (58). The results of these trials are summarised in Table 2.

A patient-level meta-analysis of 5 trials investigating shorter adjuvant trastuzumab treatment found that 6 months of treatment with trastuzumab is non-inferior to 12 months, but 9 weeks is not (59).

Escalation of adjuvant anti-HER2 therapy has also been evaluated in patients with higher-risk disease. As previously discussed, the APHINITY trial showed that patients with HER2-positive, node-positive disease benefited from the addition of pertuzumab to trastuzumab in the adjuvant setting (3-year iDFS of 92% vs 90.2%, HR 0.77; 95% CI, 0.62 to 0.96; P=0.02) (43). Extended adjuvant therapy with neratinib, a tyrosine kinase inhibitor, after trastuzumab therapy was investigated in the phase 3 ExteNet trial. This trial showed a benefit in 5-year iDFS of 90.2% in patients receiving neratinib, compared with 87.7% of those receiving the placebo (HR 0.73, 95% CI 0.57–0.92). Subgroup analysis revealed that in patients with HR-positive cancer a benefit of 5.1% in iDFS (HR 0.58, 95% CI 0.41–0.82) was shown. However, as patients in the ExteNET trial had neither received pertuzumab nor T-DM1, the actual benefit after current adjuvant and post-neoadjuvant targeted therapy could be smaller (60). Nevertheless, neratinib could offer an additional treatment option in patients with HR-positive disease.

## De-escalation strategies

The possibility of further therapy de-escalation in low-risk disease is currently being investigated in multiple clinical trials.

The WSG ADAPT HER+/HR- trial explored the feasibility of de-escalated neoadjuvant therapy in 134 patients with HER2-positive, HR-negative disease. In this trial, patients were randomly assigned to receive trastuzumab plus pertuzumab, either with or without paclitaxel. Remarkably high pCR rates (90.5%) were reported in the de-escalated chemotherapy arm after 12 weeks of paclitaxel plus dual HER2 blockade. Adjuvant therapy was given as per national guidelines. Interestingly, adjuvant chemotherapy could be omitted in patients achieving pCR at the physician's discretion, and 79% of patients who achieved pCR in the paclitaxel arm received no further chemotherapy (61). In May 2022, survival outcomes from the trial were published. Notably, patients who achieved pCR had a 5-year iDFS rate of 98%, regardless of whether they received neoadjuvant paclitaxel or were in the chemotherapy-free arm (37). While this trial is not powered to prove non-inferiority of a chemotherapy-sparing approach, these results pave the way for larger randomized control trials designed to specifically investigate whether the omission of chemotherapy may be feasible in carefully pre-selected patients.

The trial of patients with HR-positive disease, ADAPT-TP HER2 +/HR+, found that patients given neoadjuvant T-DM1 alone or with endocrine therapy were significantly more likely to achieve pCR than those given trastuzumab with endocrine therapy (41%/41.5% vs 15.1%, p<0.001). Survival data from ADAPT-TP revealed that patients who achieved pCR had similar 5-year DFS rates, regardless of whether they received chemotherapy (92.1% (95%-CI: 78-97%)

TABLE 2 Trials investigating the duration of anti-HER2 therapy.

| Trial | Phase | Duration of trastuzumab | N | Efficacy versus 1-year trastuzumab | Cardiac events |
|---|---|---|---|---|---|
| HERA (48) | III | 2 years | 5099 | DFS (10y) 70% vs 72% | 7.3% vs 4.4% |
| PHARE (53) | III | 6 months | 3380 | DFS (3y) 87.7% vs 90.7% | 1.9% vs 5.7% |
| HORG (54) | III | 6 months | 481 | DFS (3y) 93.3% vs 95.7% | 0.8% vs 0% |
| PERSEPHONE (55) | III | 6 months | 4088 | DFS (4y) 89.4% vs 89.8% | 8% vs 11% |
| SOLD (56) | III | 9 weeks | 2174 | DFS (5y) 88% vs 90.5% | 2% vs 4% |
| ShortHER (58) | III | 9 weeks | 1253 | DFS (5y) 85% vs 88% | 4.3% vs 13.1% |

with adjuvant chemotherapy vs 93% (84-97%) without adjuvant chemotherapy) (62).

As a result of these trials, further de-escalation trials were designed to prevent over-treatment. The CompassHER2-pCR and DESCRESCENDO trials are ongoing and aim to individualize adjuvant therapy based on pCR status after a de-escalated neoadjuvant course of 12 weeks paclitaxel with trastuzumab plus pertuzumab (63, 64). ATEMPT 2.0 is a phase 2 trial comparing adjuvant T-DM1 followed by trastuzumab to paclitaxel and trastuzumab, followed by trastuzumab alone. It aims to evaluate where the T-DM1 arm will have less toxicity and improved outcomes (65).

Furthermore, the omission of surgery is currently being investigated in low-risk HER2-positive early breast cancer patients who achieve a complete response to neoadjuvant therapy. In the ELPIS trial, if a complete response of the tumor is reported on the post-neoadjuvant therapy breast MRI, a vacuum-assisted breast biopsy (VAC) is performed. If on VAC no invasive or *in situ* disease is found, patients will be eligible to omit loco-regional surgery. They will instead proceed to have whole breast radiotherapy and complete 1 year of trastuzumab and pertuzumab (66).

## Biomarkers

The next challenge to enable further individualization of neoadjuvant treatment in HER2-positive breast cancer is the development of a robust biomarker to predict pCR. This would allow for the adjustment of neoadjuvant therapy by identifying patients with an increased likelihood of achieving pCR based on favorable predictive biomarkers, and identifying patients with an exceptional response to neoadjuvant therapy, who may be candidates for the omission of surgery altogether. To date, no biomarker has been validated and current recommendations are that biomarkers should not be used for monitoring patients receiving neoadjuvant therapy (67). Further research is ongoing to develop and validate potential biomarkers.

HER2-enriched intrinsic subtype, a tissue-based biomarker, has been linked with high pCR rates following neoadjuvant therapy (68). Retrospective analyses of the NOAH (69), NeoALTTO (70), CALGB40601 (71) and CHER-LOB (72) trials reported the HER2-enriched subtype to have an increased likelihood of achieving pCR with neoadjuvant chemotherapy and anti-HER2 therapy compared to other subtypes. A combined analysis of the PAMELA and TBCRC006/023 trials demonstrated that combining HER2-enriched subtype and ERBB2 mRNA levels has better sensitivity than each variable alone in predicting pCR in chemotherapy-sparing regimens (73).

Several studies have investigated tumor-infiltrating lymphocytes (TILs) as another potential biomarker for the prediction of pCR following neoadjuvant therapy in HER2-positive breast cancer. One meta-analysis reported that, regardless of the anti-HER2 agents and chemotherapy used, higher baseline TILs were associated with increased likelihood of achieving pCR (74). The PAMELA trial investigated the association between TILs and pCR in patients treated with trastuzumab and lapatinib. This study found that the presence of on-treatment TILs in HER2-positive breast cancer, measured on day 15 of treatment, was significantly associated with pCR (75). Further studies are needed to validate TILs as an accurate biomarker before it can be considered for use in clinical practice.

A pooled analysis of five prospective trials reported that PIK3CA mutant tumors significantly decreased pCR rates in HER2-positive breast cancer, particularly in HR-positive tumors (76). However, biomarker analysis of the NeoSphere study reported a non-significant decrease in pCR in patients with mutated PIK3CA (77). Therefore, PIK3CA warrants further investigation before it can be considered a potential biomarker for predicting pCR in these patients.

Blood-based biomarkers, such as circulating tumor cells (CTCs) and circulating tumor DNA (ctDNA), have also been investigated as potential predictors of pCR. One meta-analysis reported that detection of CTCs before starting neoadjuvant therapy for breast cancer was associated with a slightly lower rate of pCR (78), however, further evidence is needed to validate this. A sub-study of the NeoALTTO trial found that ctDNA detection before neoadjuvant anti-HER2 therapy was associated with decreased pCR rates (79). ctDNA detection after completion of neoadjuvant therapy has also been shown to be associated with residual disease (80–82).

Lastly, imaging-based biomarkers are also being explored as predictors of response to treatment. The use of fluorodeoxyglucose positron emission tomography (FDG-PET) as a biomarker was evaluated in the NeoALTTO (83), PHERGain (39) and TBCRC026 (84) trials. These studies suggest that these imaging strategies could facilitate further tailoring of therapy, although such strategies will require additional clinical investigation.

## Future perspectives

With substantially improved outcomes associated with the development of HER2-targeted therapies in recent years, several novel HER2-directed agents are currently being investigated in clinical trials, with promising results.

## Trastuzumab deruxtecan

Trastuzumab deruxtecan (T-DXd) is an antibody-drug conjugate, which is composed of a monoclonal antibody targeting HER2, a cleavable tetrapeptide-based linker and a topoisomerase I inhibitor (85). It has a significantly higher drug-to-antibody ratio than other antibody-drug conjugates, however the stability of the linker seems to allow for high efficacy without significant side effects. The cytotoxic payload, deruxtecan, is cell membrane permeable, giving the drug its bystander-killing effect (86).

T-DXd has shown promising results in HER2-positive breast cancer patients in the metastatic setting. In the DESTINY-Breast 01 trial, T-DXd showed a substantial benefit in patients with HER2-positive metastatic breast cancer who had previously received treatment with T-DM1 (87). Significantly improved overall response rate (ORR) and progression-free survival (PFS) was reported with T-DXd compared to T-DM1 in HER2-positive metastatic breast cancer treated with trastuzumab and a taxane in the DESTINY-Breast 03 trial (88). More recently, in the DESTINY-

Breast 04 trial involving patients with HER2-low metastatic breast cancer, treatment with T-DXd resulted in significantly longer PFS and OS than the physician's choice of chemotherapy (89).

Given the promising results of T-DXd in HER2-positive breast cancer in the metastatic setting, adjuvant and neoadjuvant T-DXd is currently under investigation. The ongoing DESTINY-Breast 05 trial is investigating T-DXd in high-risk HER2-positive disease with residual invasive breast cancer following neo-adjuvant therapy, compared to T-DM1 (90). Neoadjuvant T-DXd is also being evaluated in locally advanced or inflammatory HER2-positive breast cancer patients in the ongoing DESTINY-Breast 11 trial. This trial will compare T-DXd, alone or followed by docetaxel, trastuzumab and pertuzumab, to the current standard of care regimen (ddAC-THP) (91). The SHAMROCK study is another trial of neoadjuvant T-DXd in early stage HER2-positive breast cancer, which incorporates therapy escalation and de-escalation strategies using an on-treatment biopsy and imaging (92).

## Other novel agents

Tucatinib, a potent and selective tyrosine kinase inhibitor of HER2, is another promising agent. Tucatinib was added to trastuzumab and capecitabine in the HER2CLIMB study, resulting in improved PFS and OS in heavily pre-treated metastatic HER2-positive breast cancer (93). These results led to the design of the HER2CLIMB-05 trial, which will investigate the addition of tucatinib to standard of care maintenance in the first line setting for patients with HER2-positive metastatic breast cancer (94). Adjuvant tucatinib, in combination with T-DM1, is currently being evaluated in patients with residual disease following neo-adjuvant therapy in the COMPASS HER2 RD trial (95).

Several immune checkpoint inhibitors have been investigated in combination with HER2-directed therapies in patients with metastatic disease. Subgroup analyses from the PANACEA trial, which investigated treatment with pembrolizumab and trastuzumab in patients who had progressed on trastuzumab, showed that higher response rates were seen in PD-L1 positive tumors (96). Similarly, the KATE2 trial observed favorable PFS with atezolizumab in the subgroup of patients with PD-L1 positive tumors (97). Atezolizumab, a PD-L1 inhibitor, is being evaluated in the adjuvant setting in combination with T-DM1 in patients with residual disease after neoadjuvant therapy (98). Recently, neoadjuvant atezolizumab was investigated with docetaxel, trastuzumab and pertuzumab in HER2-positive early breast cancer and reported an acceptable pCR rate and modest toxic effects (99). Further trials on neoadjuvant immunotherapy in early HER2-positive breast cancer underway, such as NeoHIP (100) and APTneo (101) studies are underway.

## Conclusion

The introduction of HER2-directed therapies perioperatively has revolutionized the treatment of patients with HER2-positive early breast cancer. Neoadjuvant chemotherapy in combination with trastuzumab and pertuzumab has led to increased pCR rates, which in turn has significantly improved outcomes in these patients. Pathological response status provides an important guide for the appropriate adjuvant systemic therapy. De-escalation strategies are currently being investigated to avoid over treatment, and aim to safely reduce chemotherapy, while optimizing HER2-targeted therapies. The development and validation of a reliable biomarker is essential to enable these de-escalation strategies and personalization of treatment. In addition, promising novel therapies are currently being explored to further improve outcomes in HER2-positive breast cancer.

## Author contributions

Conceptualisation – AH, BH, GPD. Literature research – GPD. Manuscript preparation – GPD and SK. Manuscript review – GPD, SK, ST, GRD, BH, and AH. All authors contributed to the article and approved the submitted version.

## Conflict of interest

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

## Publisher's note



## References

1. Slamon DJ, Clark GM, Wong SG, Levin WJ, Ullrich A, McGuire WL. Human breast cancer: correlation of relapse and survival with amplification of the HER-2/neu oncogene. *Science* (1987) 235(4785):177–82. doi: 10.1126/science.3798106

2. Hurvitz SA, Hu Y, O'Brien N, Finn RS. Current approaches and future directions in the treatment of HER2-positive breast cancer. *Cancer Treat Rev* (2013) 39(3):219–29. doi: 10.1016/j.ctrv.2012.04.008

3. Keelan S, Flanagan M, Hill ADK. Evolving trends in surgical management of breast cancer: An analysis of 30 years of practice changing papers. *Front Oncol* (2021) 11:622621. doi: 10.3389/fonc.2021.622621

4. Fisher B, Bryant J, Wolmark N, Mamounas E, Brown A, Fisher ER, et al. Effect of preoperative chemotherapy on the outcome of women with operable breast cancer. *J Clin Oncol* (1998) 16(8):2672–85. doi: 10.1200/JCO.1998.16.8.2672

5. Mauri D, Pavlidis N, Ioannidis JP. Neoadjuvant versus adjuvant systemic treatment in breast cancer: A meta-analysis. *J Natl Cancer Inst* (2005) 97(3):188–94. doi: 10.1093/jnci/dji021

6. *NCCN clinical practice guidelines in oncology, breast cancer, version 2.2022–20 December 2021* (2022) (Accessed 19 July 2022). NCCN.org.

7. Giordano SH, Franzoi MAB, Temin S, Anders CK, Chandarlapaty S, Crews JR, et al. Systemic therapy for advanced human epidermal growth factor receptor 2–positive breast cancer: ASCO guideline update. *J Clin Oncol* (2022) 40(23):2612–35. doi: 10.1200/JCO.22.00519

8. *Pathological complete response in neoadjuvant treatment of high-risk early-stage breast cancer: use as an endpoint to support accelerated approval*. Silver Spring, MD: United States Food and Drug Administration, Oncology Center of Excellence (2020).

9. Cortazar P, Zhang L, Untch M, Mehta K, Costantino JP, Wolmark N, et al. Pathological complete response and long-term clinical benefit in breast cancer: the CTNeoBC pooled analysis. *Lancet* (2014) 384(9938):164–72. doi: 10.1016/S0140-6736(13)62422-8

10. Broglio KR, Quintana M, Foster M, Olinger M, McGlothlin A, Berry SM, et al. Association of pathologic complete response to neoadjuvant therapy in HER2-positive breast cancer with long-term outcomes: A meta-analysis. *JAMA Oncol* (2016) 2(6):751–60. doi: 10.1001/jamaoncol.2015.6113

11. Guarneri V, Griguolo G, Miglietta F, Conte PF, Dieci MV, Girardi F. Survival after neoadjuvant therapy with trastuzumab-lapatinib and chemotherapy in patients with HER2-positive early breast cancer: A meta-analysis of randomized trials. *ESMO Open* (2022) 7(2):100433. doi: 10.1016/j.esmoop.2022.100433

12. Davey MG, Browne F, Miller N, Lowery AJ, Kerin MJ. Pathological complete response as a surrogate to improved survival in human epidermal growth factor receptor-2-positive breast cancer: Systematic review and meta-analysis. *BJS Open* (2022) 6(3). doi: 10.1093/bjsopen/zrac028

13. Vu T, Claret FX. Trastuzumab: updated mechanisms of action and resistance in breast cancer. *Front Oncol* (2012) 2:62. doi: 10.3389/fonc.2012.00062

14. Gianni L, Eiermann W, Semiglazov V, Lluch A, Tjulandin S, Zambetti M, et al. Neoadjuvant and adjuvant trastuzumab in patients with HER2-positive locally advanced breast cancer (NOAH): Follow-up of a randomised controlled superiority trial with a parallel HER2-negative cohort. *Lancet Oncol* (2014) 15(6):640–7. doi: 10.1016/S1470-2045(14)70080-4

15. Untch M, Rezai M, Loibl S, Fasching PA, Huober J, Tesch H, et al. Neoadjuvant treatment with trastuzumab in HER2-positive breast cancer: Results from the GeparQuattro study. *J Clin Oncol* (2010) 28(12):2024–31. doi: 10.1200/JCO.2009.23.8451

16. Untch M, Fasching PA, Konecny GE, Hasmüller S, Lebeau A, Kreienberg R, et al. Pathologic complete response after neoadjuvant chemotherapy plus trastuzumab predicts favorable survival in human epidermal growth factor receptor 2-overexpressing breast cancer: results from the TECHNO trial of the AGO and GBG study groups. *J Clin Oncol* (2011) 29(25):3351–7. doi: 10.1200/JCO.2010.31.4930

17. Nami B, Maadi H, Wang Z. Mechanisms underlying the action and synergism of trastuzumab and pertuzumab in targeting HER2-positive breast cancer. *Cancers (Basel)* (2018) 10(10):342. doi: 10.3390/cancers10100342

18. Swain SM, Baselga J, Kim S-B, Ro J, Semiglazov V, Campone M, et al. Pertuzumab, trastuzumab, and docetaxel in HER2-positive metastatic breast cancer. *N Engl J Med* (2015) 372(8):724–34. doi: 10.1056/NEJMoa1413513

19. Liu X, Fang Y, Li Y, Li Y, Qi L, Wang X. Pertuzumab combined with trastuzumab compared to trastuzumab in the treatment of HER2-positive breast cancer: A systematic review and meta-analysis of randomized controlled trials. *Front Oncol* (2022) 12:894861. doi: 10.3389/fonc.2022.894861

20. Segovia-Mendoza M, González-González ME, Barrera D, Díaz L, García-Becerra R. Efficacy and mechanism of action of the tyrosine kinase inhibitors gefitinib, lapatinib and neratinib in the treatment of HER2-positive breast cancer: preclinical and clinical evidence. *Am J Cancer Res* (2015) 5(9):2531–61.

21. Blackwell KL, Burstein HJ, Storniolo AM, Rugo HS, Sledge G, Aktan G, et al. Overall survival benefit with lapatinib in combination with trastuzumab for patients with human epidermal growth factor receptor 2-positive metastatic breast cancer: Final results from the EGF104900 study. *J Clin Oncol* (2012) 30(21):2585–92. doi: 10.1200/JCO.2011.35.6725

22. Guarneri V, Dieci MV, Griguolo G, Miglietta F, Girardi F, Bisagni G, et al. Trastuzumab-lapatinib as neoadjuvant therapy for HER2-positive early breast cancer: Survival analyses of the CHER-lob trial. *Eur J Cancer* (2021) 153:133–41. doi: 10.1016/j.ejca.2021.05.018

23. Fernandez-Martinez A, Krop IE, Hillman DW, Polley M-Y, Parker JS, Huebner L, et al. Survival, pathologic response, and genomics in CALGB 40601 (Alliance), a neoadjuvant phase III trial of paclitaxel-trastuzumab with or without lapatinib in HER2-positive breast cancer. *J Clin Oncol* (2020) 38(35):4184–93. doi: 10.1200/JCO.20.01276

24. Huober J, Holmes E, Baselga J, de Azambuja E, Untch M, Fumagalli D, et al. Survival outcomes of the NeoALTTO study (BIG 1-06): updated results of a randomised multicenter phase III neoadjuvant clinical trial in patients with HER2-positive primary breast cancer. *Eur J Cancer* (2019) 118:169–77. doi: 10.1016/j.ejca.2019.04.038

25. Robidoux A, Tang G, Rastogi P, Geyer CE, Azar CA, Atkins JN, et al. Evaluation of lapatinib as a component of neoadjuvant therapy for HER2+ operable breast cancer: 5-year outcomes of NSABP protocol b-41. *J Clin Oncol* (2016) 34(15_suppl):501. doi: 10.1200/JCO.2016.34.15_suppl.501

26. Bundred N, Porta N, Brunt AM, Cramer A, Hanby A, Shaaban AM, et al. Combined perioperative lapatinib and trastuzumab in early HER2-positive breast cancer identifies early responders: Randomized UK EPHOS-b trial long-term results. *Clin Cancer Res* (2022) 28(7):1323–34. doi: 10.1158/1078-0432.CCR-21-3177

27. Gunasekara ADM, Anothaisintawee T, Youngkong S, Ha NT, McKay GJ, Attia J, et al. Neoadjuvant treatment with HER2-targeted therapies in HER2-positive breast cancer: A systematic review and network meta-analysis. *Cancers (Basel)* (2022) 14(3):523. doi: 10.3390/cancers14030523

28. van der Voort A, van Ramshorst MS, van Werkhoven ED, Mandjes IA, Kemper I, Vulink AJ, et al. Three-year follow-up of neoadjuvant chemotherapy with or without anthracyclines in the presence of dual ERBB2 blockade in patients with ERBB2-positive breast cancer: A secondary analysis of the TRAIN-2 randomized, phase 3 trial. *JAMA Oncol* (2021) 7(7):978–84. doi: 10.1001/jamaoncol.2021.1371

29. Schneeweiss A, Chia S, Hickish T, Harvey V, Eniu A, Waldron-Lynch M, et al. Long-term efficacy analysis of the randomised, phase II TRYPHAENA cardiac safety study: Evaluating pertuzumab and trastuzumab plus standard neoadjuvant anthracycline-containing and anthracycline-free chemotherapy regimens in patients with HER2-positive early breast cancer. *Eur J Cancer* (2018) 89:27–35. doi: 10.1016/j.ejca.2017.10.021

30. Swain SM, Ewer MS, Viale G, Delaloge S, Ferrero JM, Verrill M, et al. Pertuzumab, trastuzumab, and standard anthracycline- and taxane-based chemotherapy for the neoadjuvant treatment of patients with HER2-positive localized breast cancer (BERENICE): a phase II, open-label, multicenter, multinational cardiac safety study. *Ann Oncol* (2018) 29(3):646–53. doi: 10.1093/annonc/mdx773

31. Dang C, Ewer MS, Delaloge S, Ferrero JM, Colomer R, de la Cruz-Merino L, et al. BERENICE final analysis: Cardiac safety study of neoadjuvant pertuzumab, trastuzumab, and chemotherapy followed by adjuvant pertuzumab and trastuzumab in HER2-positive early breast cancer. *Cancers (Basel)* (2022) 14(11):2596. doi: 10.3390/cancers14112596

32. Hurvitz SA, Martin M, Jung KH, Huang C-S, Harbeck N, Valero V, et al. Neoadjuvant trastuzumab emtansine and pertuzumab in human epidermal growth factor receptor 2–positive breast cancer: Three-year outcomes from the phase III KRISTINE study. *J Clin Oncol* (2019) 37(25):2206–16. doi: 10.1200/JCO.19.00882

33. Robidoux A, Tang G, Rastogi P, Geyer CEJr., Azar CA, Atkins JN, et al. Lapatinib as a component of neoadjuvant therapy for HER2-positive operable breast cancer (NSABP protocol b-41): an open-label, randomised phase 3 trial. *Lancet Oncol* (2013) 14(12):1183–92. doi: 10.1016/S1470-2045(13)70411-X

34. Untch M, von Minckwitz G, Gerber B, Schem C, Rezai M, Fasching PA, et al. Survival analysis after neoadjuvant chemotherapy with trastuzumab or lapatinib in patients with human epidermal growth factor receptor 2-positive breast cancer in the GeparQuinto (G5) study (GBG 44). *J Clin Oncol* (2018) 36(13):1308–16. doi: 10.1200/JCO.2017.75.9175

35. Gianni L, Pienkowski T, Im Y-H, Tseng L-M, Liu M-C, Lluch A, et al. 5-year analysis of neoadjuvant pertuzumab and trastuzumab in patients with locally advanced, inflammatory, or early-stage HER2-positive breast cancer (NeoSphere): a multicentre, open-label, phase 2 randomised trial. *Lancet Oncol* (2016) 17(6):791–800. doi: 10.1016/S1470-2045(16)00163-7

36. Untch M, Jackisch C, Schneeweiss A, Schmatloch S, Aktas B, Denkert C, et al. NAB-paclitaxel improves disease-free survival in early breast cancer: GBG 69–GeparSepto. *J Clin Oncol* (2019) 37(25):2226–34. doi: 10.1200/JCO.18.01842

37. Nitz U, Gluz O, Graeser M, Christgen M, Kuemmel S, Grischke E-M, et al. De-escalated neoadjuvant pertuzumab plus trastuzumab therapy with or without weekly paclitaxel in HER2-positive, hormone receptor-negative, early breast cancer (WSG-ADAPT-HER2+/HR-): survival outcomes from a multicentre, open-label, randomised, phase 2 trial. *Lancet Oncol* (2022) 23(5):625–35. doi: 10.1016/S1470-2045(22)00159-0

38. Dang C, Ewer MS, Delaloge S, Ferrero JM, Colomer R, de la Cruz Merino L, et al. 43O pertuzumab/trastuzumab in early stage HER2-positive breast cancer: 5-year and final analysis of the BERENICE trial. *Ann Oncol* (2021) 32:S38–S9. doi: 10.1016/j.annonc.2021.03.057

39. Pérez-García JM, Gebhart G, Ruiz Borrego M, Stradella A, Bermejo B, Schmid P, et al. Chemotherapy de-escalation using an 18F-FDG-PET-based pathological response-adapted strategy in patients with HER2-positive early breast cancer (PHERGain): a multicentre, randomised, open-label, non-comparative, phase 2 trial. *Lancet Oncol* (2021) 22(6):858–71. doi: 10.1016/S1470-2045(21)00122-4

40. Hatschek T, Foukakis T, Björhle J, Lekberg T, Fredholm H, Elinder E, et al. Neoadjuvant trastuzumab, pertuzumab, and docetaxel vs trastuzumab emtansine in patients with ERBB2-positive breast cancer: A phase 2 randomized clinical trial. *JAMA Oncol* (2021) 7(9):1360–7. doi: 10.1001/jamaoncol.2021.1932

41. Tolaney SM, Guo H, Pernas S, Barry WT, Dillon DA, Ritterhouse L, et al. Seven-year follow-up analysis of adjuvant paclitaxel and trastuzumab trial for node-negative, human epidermal growth factor receptor 2–positive breast cancer. *J Clin Oncol* (2019) 37(22):1868–75. doi: 10.1200/JCO.19.00066

42. Ditsch N, Kolberg-Liedtke C, Friedrich M, Jackisch C, Albert US, Banys-Paluchowski M, et al. AGO recommendations for the diagnosis and treatment of patients with early breast cancer: Update 2021. *Breast Care* (2021) 16(3):214–27. doi: 10.1159/000516419

43. Piccart M, Procter M, Fumagalli D, Azambuja Ed, Clark E, Ewer MS, et al. Adjuvant pertuzumab and trastuzumab in early HER2-positive breast cancer in the APHINITY trial: 6 years' follow-up. *J Clin Oncol* (2021) 39(13):1448–57. doi: 10.1200/JCO.20.01204

44. Loibl S, Jassem J, Sonnenblick A, Parlier D, Winer E, Bergh J, et al. VP6-2022: Adjuvant pertuzumab and trastuzumab in patients with early HER-2 positive breast cancer in APHINITY: 8. *4 years' follow-up Ann Oncol* (2022) 33(9):986–7. doi: 10.1016/j.annonc.2022.06.009

45. von Minckwitz G, Huang C-S, Mano MS, Loibl S, Mamounas EP, Untch M, et al. Trastuzumab emtansine for residual invasive HER2-positive breast cancer. *New Engl J Med* (2018) 380(7):617–28. doi: 10.1056/NEJMoa1814017

46. Tolaney SM, Tayob N, Dang C, Yardley DA, Isakoff SJ, Valero V, et al. Adjuvant trastuzumab emtansine versus paclitaxel in combination with trastuzumab for stage I HER2-positive breast cancer (ATEMPT): A randomized clinical trial. *J Clin Oncol* (2021) 39(21):2375–85. doi: 10.1200/JCO.20.03398

47. Krop IE, Im S-A, Barrios C, Bonnefoi H, Gralow J, Toi M, et al. Trastuzumab emtansine plus pertuzumab versus taxane plus trastuzumab plus pertuzumab after anthracycline for high-risk human epidermal growth factor receptor 2–positive early breast cancer: The phase III KAITLIN study. *J Clin Oncol* (2022) 40(5):438–48. doi: 10.1200/JCO.21.00896

48. Cameron D, Piccart-Gebhart MJ, Gelber RD, Procter M, Goldhirsch A, de Azambuja E, et al. 11 years' follow-up of trastuzumab after adjuvant chemotherapy in HER2-positive early breast cancer: final analysis of the HERceptin adjuvant (HERA) trial. *Lancet* (2017) 389(10075):1195–205. doi: 10.1016/S0140-6736(16)32616-2

49. Perez EA, Romond EH, Suman VJ, Jeong J-H, Sledge G, Geyer CEJr, et al. Trastuzumab plus adjuvant chemotherapy for human epidermal growth factor receptor 2–positive breast cancer: Planned joint analysis of overall survival from NSABP b-31 and NCCTG N9831. *J Clin Oncol* (2014) 32(33):3744–52. doi: 10.1200/JCO.2014.55.5730

50. Romond EH, Perez EA, Bryant J, Suman VJ, Geyer CEJr., Davidson NE, et al. Trastuzumab plus adjuvant chemotherapy for operable HER2-positive breast cancer. *N Engl J Med* (2005) 353(16):1673–84. doi: 10.1056/NEJMoa052122

51. Slamon D, Eiermann W, Robert N, Pienkowski T, Martin M, Press M, et al. Adjuvant trastuzumab in HER2-positive breast cancer. *N Engl J Med* (2011) 365 (14):1273–83. doi: 10.1056/NEJMoa0910383

52. Bradley R, Braybrooke J, Gray R, Hills R, Liu Z, Peto R, et al. Trastuzumab for early-stage, HER2-positive breast cancer: A meta-analysis of 13 864 women in seven randomised trials. *Lancet Oncol* (2021) 22(8):1139–50. doi: 10.1016/S1470-2045(21) 00288-6

53. Pivot X, Romieu G, Debled M, Pierga J-Y, Kerbrat P, Bachelot T, et al. 6 months versus 12 months of adjuvant trastuzumab in early breast cancer (PHARE): Final analysis of a multicentre, open-label, phase 3 randomised trial. *Lancet* (2019) 393(10191):2591–8. doi: 10.1016/S0140-6736(19)30653-1

54. Mavroudis D, Saloustros E, Malamos N, Kakolyris S, Boukovinas I, Papakotoulas P, et al. Six versus 12 months of adjuvant trastuzumab in combination with dose-dense chemotherapy for women with HER2-positive breast cancer: a multicenter randomized study by the Hellenic oncology research group (HORG). *Ann Oncol* (2015) 26(7):1333–40. doi: 10.1093/annonc/mdv213

55. Earl HM, Hiller L, Vallier AL, Loi S, McAdam K, Hughes-Davies L, et al. 6 versus 12 months of adjuvant trastuzumab for HER2-positive early breast cancer (PERSEPHONE): 4-year disease-free survival results of a randomised phase 3 non-inferiority trial. *Lancet* (2019) 393 (10191):2599–612. doi: 10.1016/S0140-6736(19)30650-6

56. Joensuu H, Fraser J, Wildiers H, Huovinen R, Auvinen P, Utriainen M, et al. Effect of adjuvant trastuzumab for a duration of 9 weeks vs 1 year with concomitant chemotherapy for early human epidermal growth factor receptor 2–positive breast cancer: The SOLD randomized clinical trial. *JAMA Oncol* (2018) 4(9):1199–206. doi: 10.1001/jamaoncol.2018.1380

57. Conte P, Frassoldati A, Bisagni G, Brandes AA, Donadio M, Garrone O, et al. Nine weeks versus 1 year adjuvant trastuzumab in combination with chemotherapy: final results of the phase III randomized short-HER study‡. *Ann Oncol* (2018) 29(12):2328–33. doi: 10.1093/annonc/mdy414

58. Conte P, Frassoldati A, Bisagni G, Brandes AA, Donadio M, Garrone O, et al. 41O nine weeks vs 1-year adjuvant trastuzumab: Long term outcomes of the ShortHER randomised trial. *Ann Oncol* (2021) 32:S37. doi: 10.1016/j.annonc.2021.03.055

59. Earl HM, Hiller L, Dunn JA, Conte P, D'Amico R, Guarneri V, et al. LBA11 individual patient data meta-analysis of 5 non-inferiority RCTs of reduced duration single agent adjuvant trastuzumab in the treatment of HER2 positive early breast cancer. *Ann Oncol* (2021) 32:S1283. doi: 10.1016/j.annonc.2021.08.2083

60. Chan A, Moy B, Mansi J, Ejlertsen B, Holmes FA, Chia S, et al. Final efficacy results of neratinib in HER2-positive hormone receptor-positive early-stage breast cancer from the phase III ExteNET trial. *Clin Breast Cancer* (2021) 21(1):80–91.e7. doi: 10.1016/j.clbc.2020.09.014

61. Nitz UA, Gluz O, Christgen M, Grischke EM, Augustin D, Kuemmel S, et al. De-escalation strategies in HER2-positive early breast cancer (EBC): final analysis of the WSG-ADAPT HER2+/HR-; phase II trial: efficacy, safety, and predictive markers for 12 weeks of neoadjuvant dual blockade with trastuzumab and pertuzumab ± weekly paclitaxel. *Ann Oncol* (2017) 28(11):2768–72. doi: 10.1093/annonc/mdx494

62. Gluz O, Nitz U, Christgen M, Kuemmel S, Holtschmidt J, Priel J, et al. De-escalated chemotherapy versus endocrine therapy plus pertuzumab+ trastuzumab for HR+/HER2+ early breast cancer (BC): First efficacy results from the neoadjuvant WSG-TP-II study. *J Clin Oncol* (2020) 38(15_suppl):515–. doi: 10.1200/JCO.2020.38.15_suppl.515

63. *CompassHER2-pCR: Decreasing chemotherapy for breast cancer patients after pre-surgery chemo and targeted therapy*. Available at: https://ClinicalTrials.gov/show/NCT04266249.

64. *De-escalation adjuvant chemo in HER2+/ER-/Node-neg early BC patients who achieved pCR after neoadjuvant chemo & dual HER2 blockade*. Available at: https://ClinicalTrials.gov/show/NCT04675827.

65. *ATEMPT 2.0: Adjuvant T-DM1 vs TH*. Available at: https://ClinicalTrials.gov/show/NCT04893109.

66. Pascual T, Chic N, Martinez Saez O, Sanfeliu Torres E, Adamo B, Cebrecos I, et al. 132TiP HCB-ONC001 ELPIS TRIAL: Omission of surgery and sentinel lymph node dissection in clinically low-risk HER2-positive breast cancer with high HER2 addiction and a complete response following standard anti-HER2-based neoadjuvant therapy. *Ann Oncol* (2022) 33:S182–3. doi: 10.1016/j.annonc.2022.03.149

67. Korde LA, Somerfield MR, Carey LA, Crews JR, Denduluri N, Hwang ES, et al. Neoadjuvant chemotherapy, endocrine therapy, and targeted therapy for breast cancer: ASCO guideline. *J Clin Oncol* (2021) 39(13):1485–505. doi: 10.1200/JCO.20.03399

68. Schettini F, Pascual T, Conte B, Chic N, Brasó-Maristany F, Galván P, et al. HER2-enriched subtype and pathological complete response in HER2-positive breast cancer: A systematic review and meta-analysis. *Cancer Treat Rev* (2020) 84:101965. doi: 10.1016/j.ctrv.2020.101965

69. Prat A, Bianchini G, Thomas M, Belousov A, Cheang MC, Koehler A, et al. Research-based PAM50 subtype predictor identifies higher responses and improved survival outcomes in HER2-positive breast cancer in the NOAH study. *Clin Cancer Res* (2014) 20(2):511–21. doi: 10.1158/1078-0432.CCR-13-0239

70. Fumagalli D, Venet D, Ignatiadis M, Azim HAJr., Maetens M, Rothé F, et al. RNA Sequencing to predict response to neoadjuvant anti-HER2 therapy: A secondary analysis of the NeoALTTO randomized clinical trial. *JAMA Oncol* (2017) 3(2):227–34. doi: 10.1001/jamaoncol.2016.3824

71. Carey LA, Berry DA, Cirrincione CT, Barry WT, Pitcher BN, Harris LN, et al. Molecular heterogeneity and response to neoadjuvant human epidermal growth factor receptor 2 targeting in CALGB 40601, a randomized phase III trial of paclitaxel plus trastuzumab with or without lapatinib. *J Clin Oncol* (2016) 34(6):542–9. doi: 10.1200/JCO.2015.62.1268

72. Dieci MV, Prat A, Tagliafico E, Paré L, Ficarra G, Bisagni G, et al. Integrated evaluation of PAM50 subtypes and immune modulation of pCR in HER2-positive breast cancer patients treated with chemotherapy and HER2-targeted agents in the CherLOB trial. *Ann Oncol* (2016) 27(10):1867–73. doi: 10.1093/annonc/mdw262

73. Prat A, Pascual T, De Angelis C, Gutierrez C, Llombart-Cussac A, Wang T, et al. HER2-enriched subtype and ERBB2 expression in HER2-positive breast cancer treated with dual HER2 blockade. *J Natl Cancer Inst* (2020) 112(1):46–54. doi: 10.1093/jnci/djz042

74. Solinas C, Ceppi M, Lambertini M, Scartozzi M, Buisseret L, Garaud S, et al. Tumor-infiltrating lymphocytes in patients with HER2-positive breast cancer treated with neoadjuvant chemotherapy plus trastuzumab, lapatinib or their combination: A meta-analysis of randomized controlled trials. *Cancer Treat Rev* (2017) 57:8–15. doi: 10.1016/j.ctrv.2017.04.005

75. Nuciforo P, Prat A, Llombart A, Fasani R, Paré L, Pascual T, et al. Tumor-infiltrating lymphocytes (TILs) in HER2-positive (HER2+) early breast cancer treated with neoadjuvant lapatinib and trastuzumab without chemotherapy in the PAMELA trial. *Ann Oncol* (2017) 28:v46. doi: 10.1093/annonc/mdx362.006

76. Loibl S, Majewski I, Guarneri V, Nekljudova V, Holmes E, Bria E, et al. PIK3CA mutations are associated with reduced pathological complete response rates in primary HER2-positive breast cancer: pooled analysis of 967 patients from five prospective trials investigating lapatinib and trastuzumab. *Ann Oncol* (2016) 27(8):1519–25. doi: 10.1093/annonc/mdw197

77. Bianchini G, Kiermaier A, Bianchi GV, Im YH, Pienkowski T, Liu MC, et al. Biomarker analysis of the NeoSphere study: pertuzumab, trastuzumab, and docetaxel versus trastuzumab plus docetaxel, pertuzumab plus trastuzumab, or pertuzumab plus docetaxel for the neoadjuvant treatment of HER2-positive breast cancer. *Breast Cancer Res* (2017) 19(1):16. doi: 10.1186/s13058-017-0806-9

78. Bidard F-C, Michiels S, Riethdorf S, Mueller V, Esserman LJ, Lucci A, et al. Circulating tumor cells in breast cancer patients treated by neoadjuvant chemotherapy: A meta-analysis. *JNCI: J Natl Cancer Institute* (2018) 110(6):560–7. doi: 10.1093/jnci/djy018

79. Rothé F, Silva MJ, Venet D, Campbell C, Bradburry I, Rouas G, et al. Circulating tumor DNA in HER2-amplified breast cancer: A translational research substudy of the NeoALTTO phase III trial. *Clin Cancer Res* (2019) 25(12):3581–8. doi: 10.1158/1078-0432.CCR-18-2521

80. McDonald BR, Contente-Cuomo T, Sammut SJ, Odenheimer-Bergman A, Ernst B, Perdigones N, et al. Personalized circulating tumor DNA analysis to detect residual disease after neoadjuvant therapy in breast cancer. *Sci Transl Med* (2019) 11(504): eaax7392. doi: 10.1126/scitranslmed.aax7392

81. Moss J, Zick A, Grinshpun A, Carmon E, Maoz M, Ochana BL, et al. Circulating breast-derived DNA allows universal detection and monitoring of localized breast cancer. *Ann Oncol* (2020) 31(3):395–403. doi: 10.1016/j.annonc.2019.11.014

82. Ciriaco N, Zamora E, Escrivá-de-Romaní S, Miranda Gómez I, Jiménez Flores J, Saura C, et al. Clearance of ctDNA in triple-negative and HER2-positive breast cancer patients during neoadjuvant treatment is correlated with pathologic complete response. *Ther Adv Med Oncol* (2022) 14:17588359221139601. doi: 10.1177/17588359221139601

83. Di Cosimo S, Campbell C, Azim HAJr., Galli G, Bregni G, Curigliano G, et al. The use of breast imaging for predicting response to neoadjuvant lapatinib, trastuzumab and their combination in HER2-positive breast cancer: Results from neo-ALTTO. *Eur J Cancer* (2018) 89:42–8. doi: 10.1016/j.ejca.2017.10.036

84. Connolly RM, Leal JP, Solnes L, Huang CY, Carpenter A, Gaffney K, et al. Updated results of TBCRC026: Phase II trial correlating standardized uptake value with pathological complete response to pertuzumab and trastuzumab in breast cancer. *J Clin Oncol* (2021) 39(20):2247–56. doi: 10.1200/JCO.21.00280

85. Modi S, Saura C, Yamashita T, Park YH, Kim S-B, Tamura K, et al. Trastuzumab deruxtecan in previously treated HER2-positive breast cancer. *New Engl J Med* (2019) 382 (7):610–21. doi: 10.1056/NEJMoa1914510

86. Doi T, Shitara K, Naito Y, Shimomura A, Fujiwara Y, Yonemori K, et al. Safety, pharmacokinetics, and antitumour activity of trastuzumab deruxtecan (DS-8201), a HER2-targeting antibody-drug conjugate, in patients with advanced breast and gastric

or gastro-oesophageal tumours: a phase 1 dose-escalation study. *Lancet Oncol* (2017) 18 (11):1512–22. doi: 10.1016/S1470-2045(17)30604-6

87. Modi S, Saura C, Yamashita T, Park YH, Kim SB, Tamura K, et al. Trastuzumab deruxtecan in previously treated HER2-positive breast cancer. *N Engl J Med* (2020) 382 (7):610–21. doi: 10.1056/NEJMoa1914510

88. Cortés J, Kim SB, Chung WP, Im SA, Park YH, Hegg R, et al. Trastuzumab deruxtecan versus trastuzumab emtansine for breast cancer. *N Engl J Med* (2022) 386 (12):1143–54. doi: 10.1056/NEJMoa2115022

89. Modi S, Jacot W, Yamashita T, Sohn J, Vidal M, Tokunaga E, et al. Trastuzumab deruxtecan in previously treated HER2-low advanced breast cancer. *N Engl J Med* (2022) 387(1):9–20. doi: 10.1056/NEJMoa2203690

90. *A study of trastuzumab deruxtecan (T-DXd) versus trastuzumab emtansine (T-DM1) in high-risk HER2-positive participants with residual invasive breast cancer following neoadjuvant therapy (DESTINY-Breast05)*. Available at: https://ClinicalTrials.gov/show/NCT04622319.

91. *Trastuzumab deruxtecan (T-DXd) alone or in sequence with THP, versus standard treatment (ddAC-THP), in HER2-positive early breast cancer*. Available at: https://ClinicalTrials.gov/show/NCT05113251.

92. *Single arm phase 2 trial of neoadjuvant trastuzumab deruxtecan (T-DXd) with response-directed definitive therapy in early stage HER2-positive breast cancer: a standard chemotherapy-sparing approach to curative-intent treatment – SHAMROCK study*. Available at: https://eudract.ema.europa.eu/results-web/index.xhtml.

93. Curigliano G, Mueller V, Borges V, Hamilton E, Hurvitz S, Loi S, et al. Tucatinib versus placebo added to trastuzumab and capecitabine for patients with pretreated HER2 + metastatic breast cancer with and without brain metastases (HER2CLIMB): final overall survival analysis. *Ann Oncol* (2022) 33(3):321–9. doi: 10.1016/j.annonc.2021.12.005

94. *A study of tucatinib or placebo with trastuzumab and pertuzumab for metastatic HER2+ breast cancer*. Available at: https://ClinicalTrials.gov/show/NCT05132582.

95. O'Sullivan CCM, Ballman KV, McCall LM, Zemla TJ, Weiss A, Mitchell M, et al. A011801 (CompassHER2 RD): Postneoadjuvant T-DM1 + tucatinib/placebo in patients with residual HER2-positive invasive breast cancer. *J Clin Oncol* (2021) 39(15_suppl): TPS595–TPS. doi: 10.1200/JCO.2021.39.15_suppl.TPS595

96. Loi S, Giobbie-Hurder A, Gombos A, Bachelot T, Hui R, Curigliano G, et al. Pembrolizumab plus trastuzumab in trastuzumab-resistant, advanced, HER2-positive breast cancer (PANACEA): A single-arm, multicentre, phase 1b-2 trial. *Lancet Oncol* (2019) 20(3):371–82. doi: 10.1016/S1470-2045(18)30812-X

97. Emens LA, Esteva FJ, Beresford M, Saura C, De Laurentiis M, Kim SB, et al. Trastuzumab emtansine plus atezolizumab versus trastuzumab emtansine plus placebo in previously treated, HER2-positive advanced breast cancer (KATE2): A phase 2, multicentre, randomised, double-blind trial. *Lancet Oncol* (2020) 21(10):1283–95. doi: 10.1016/S1470-2045(20)30465-4

98. Hurvitz SA, Bachelot T, Bianchini G, Harbeck N, Loi S, Park YH, et al. ASTEFANIA: adjuvant ado-trastuzumab emtansine and atezolizumab for high-risk, HER2-positive breast cancer. *Future Oncol* (2022) 18(32):3563–72. doi: 10.2217/fon-2022-0485

99. Ahn HK, Sim SH, Suh KJ, Kim MH, Jeong JH, Kim J-Y, et al. Response rate and safety of a neoadjuvant pertuzumab, atezolizumab, docetaxel, and trastuzumab regimen for patients with ERBB2-positive stage II/III breast cancer: The neo-PATH phase 2 nonrandomized clinical trial. *JAMA Oncol* (2022) 8(9):1271–7. doi: 10.1001/jamaoncol.2022.2310

100. McArthur HL, Leal JHS, Page DB, Abaya CD, Basho RK, Phillips M, et al. Neoadjuvant HER2-targeted therapy +/- immunotherapy with pembrolizumab (neoHIP): An open-label randomized phase II trial. *J Clin Oncol* (2022) 40(16_suppl):TPS624–TPS. doi: 10.1200/JCO.2022.40.16_suppl.TPS624

101. *Neoadjuvant treatment of HER2 positive early high-risk and locally advanced breast cancer (APTneo)*. Available at: https://ClinicalTrials.gov/show/NCT03595592.

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# Application of Clavien–Dindo classfication-grade in evaluating overall efficacy of laparoscopic pancreaticoduodenectomy

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**Background:** The Clavien–Dindo classification (CDC) has been widely accepted and applied in clinical practice. We investigated its effectiveness in prediction of major complications (LPPC) after laparoscopic pancreaticoduodenectomy (LPD) and associated risk factors.
**Methods:** A retrospective analysis was conducted covering clinical data of 793 patients undergoing LPD from April 2015 to November 2021. CDC was utilized to grade postoperative complications and analyze the differences. Risk factors of LPPC were identified according to univariate and multivariate analyses.
**Resluts:** For the 793 patients undergoing laparoscopic pancreaticoduodenectomy in the northeast of China, LPPC was reported in 260 (32.8%) patients, pancreatic fistula in 169 (21.3%), biliary fistula in 44 (5.5%), delayed gastric emptying in 17 (2.1%), post pancreatectomy hemorrhage in 55 (6.9%), intestinal fistula in 7 (0.8%), abdominal infections in 59 (7.4%) and pulmonary complication in 28 (3.5%). All complications were classified into five levels with the C–D classification (Grade I–V), with 83 (31.9%) patients as grade I, 91 (35.0%) as grade II, 38 (14.6%) as grade IIIa, 24 (9.2%) as grade IIIb, 9 (3.5%) as grade IV and 15 (5.8%) as grade V. 86 (10.8%) patients experienced major complications (grade III–V).The results of univariate and multivariate analysis revealed the independent risk factors for laparoscopic pancreaticoduodenectomy complications to be preoperative total bilirubin ($P = 0.029$, OR = 1.523), soft pancreas texture ($P < 0.001$, OR = 1.399), male ($P = 0.038$, OR = 1.396) and intraoperative transfusion ($P = 0.033$, OR = 1.517). Preoperative total bilirubin ($P = 0.036$, OR = 1.906) and intraoperative transfusions ($P = 0.004$, OR = 2.123) were independently associated with major postoperative complications. The influence of different bilirubin levels on C–D grade of complications was statistically significant ($P = 0.036$, OR = 1.906).
**Conclusions:** The Clavien–Dindo classification (CDC) may serve as a valid tool to predict major postoperative complications and contribute to perioperative management and comparison of surgical techniques in different medical centers.

KEYWORDS
Clavien–Dindo classification, complications, surgery, laparscope, pancreaticoduodenectomy

## Introduction

Pancreaticoduodenectomy (PD), as a preferred treatment for malignant diseases of the head of pancreas, distal bile duct and periampullary. In contrast, greater advantages have been reported of laparoscopic surgery over PD (1–3). Despite the significant modifications in medical technology, the complication rates are still reported to be around 50% (4–7) in

high volume centers, which has prolonged hospital stays, bringing mental burden to patients and aggravate health care costs. Hence, the overall evaluation of surgical complications has absorbed great concern in recent years.

Over the past decade, there have appeared various definitions of postoperative complication. For instance, the international study group of pancreatic surgery (ISGPS) reported a definition of post pancreatectomy hemorrhage (PPH) (8) and postoperative pancreatic fistula(POPF) (9). Charles J (10) defined the delayed gastric emptying (DGE) which requires postoperative nasogastric tube decompression for over 10 days. However, these definitions are only rooted in a single system, without the available established criteria to standardize surgical complications. The lack of a uniform criterion involving all systemic complications impedes effective comparison of surgical outcomes and levels of practice across medical bases, resulting in inaccurate recording of major complications incidence.

In terms of the categorization of postoperative complications of LPD, ISGPS has introduced a series of definitions, which have received wide adoption and favor from domestic and international surgical groups. However, these definitions are limited to only a specific class of PPC, covering PPH, POPF and DGE, and show the unique gas assessment criteria for a specific complication, which requires the assessment and exploration on the relevant risk factors only for a specific class of complication, while inefficient for the synergy and risk factors among these different classifications of complications. Secondly, a simple, reproducible evaluation that works for all types of postoperative complications is required considering the increasing health care need and medical costs, the limited resources, and variation in clinical perioperative data, so as to achieve the long-term comparisons between medical centers, between surgical modalities, and within the same center. The C–D grading system developed by Clavien et al. provides such a new approach. Dindo proposed (11) an modified grading system referring to complication management in 2004, which has been widely adopted by surgeons around the world. The Japan Clinical Oncology Group (12) set up a committee and detailed the grading criteria based on the rules of CDC. Laura (13) utilized CDC to explore the impact of complications following minimally invasive esophagectomy on survival. Dong-Kyu (14) also evaluated complications after small bowel resection depending on CDC. While limited was known about the application of CDC to LPD. The objective of this study is to identify risk factors for LPPC and to determine their association with CDC through a retrospective analysis of the largest LPD volume center in northeast China. By evaluating the overall postoperative efficacy of LPD, we hoped to make a contribution to a personalized management of patients undergoing LPD.

## Patients and methods

All patients who underwent LPD at the First Affiliated Hospital of Jilin University from April 2015 to November 2021 were involved in this study, which was approved by the hospital. A prospective electronic database was maintained to provide all the data, containing all of the patients' outpatient and inpatient information, covering preoperative laboratory parameters (serum total bilirubin), preoperative biliary drainage, common disease (hypertension, diabetes, hepatitis), patients characteristics, preoperative surgical factors, outcomes and postoperative treatment. Considering the varying views of different surgeons on the indications for surgery, the serum albumin and hemoglobin were maintained above 35 g/L and 100 g/L, respectively, before surgery here. Therefore, these two variables were excluded from the study. The patient had signed an informed consent for the data to be used in the clinical study. The information will be maintained strictly confidential. The study was approved by the First Affiliated Hospital of Jilin University and all methods were performed in accordance with the relevant guidelines and regulations.

Patients with these identifications were not included in the model. First, preoperative enhanced computerized tomography (CT) or magnetic resonance imaging (MRI) indicate distant metastasis of malignant cells. Second, intraoperative tumors invade arteries, veins and surrounding vital organs, or extensive abdominal metastasis fails to be completely resected. Third, due to bleeding or severe tissue adhesion, intraoperative tumors are difficult to operate and switch to open.

## Surgery

All procedures following the standard of classical Whipple surgery were performed by four experienced surgeons through minimally invasive laparoscopy. Removed organs referred to the gastric pylorus, distal antrum of the stomach, duodenum, cholecyst, distal common bile duct, proximal jejunum and head of pancreas. The gastric antrum and neck of the pancreas were disconnected by endovascular gastrointestinal anastomosis stapler, without performing enlarged lymph node dissection. Digestive tract reconstruction was performed by Child method. Pancreatoenteric was performed by means of pancreatic duct anastomosis to jejunum mucosa. The remaining pancreas was routinely placed with a supportive tube to ensure the smooth drainage of pancreatic fluid. Abdominal drainage tubes were indwelled in front and rear of pancreaticoenteric anastomosis and around bilioenteric anastomosis. All patients received cefoperazone shock therapy before surgery, routine prophylactic therapy with antibiotics (Cefoperazone 1 g, BID, intravenous drip) and somatostatin (Stilamin 6 mg, QD, intravenous drip) after surgery, given hemostatic drugs to prevent bleeding.

## Complications

Case records were reviewed for each enrolled patient to identify complications, including PPH, POPF, DGE, biliary fistula, abdominal infections, pulmonary complication, and intestinal fistula. PPH,POPF,DGE and biliary fistula were defined in ISGPS standards (8, 9, 15, 16). All complications were graded (grade I–V)

per Clavien–Dindo classification. Major complications were defined as severely greater than or equal to grade III. Mortality was defined as death within 30 days after surgery or during hospitalization. Mortality is the rate of grade V complications.

The specific grading criteria are: (1) Grade I: Any deviation from the normal postoperative normal recovery process that includes only the use of antiemetics, antipyretics, analgesics without requirement of pharmacological treatment, surgical intervention, endoscopic or interventional treatment. Only those can be resolved with antiemetics, antipyretics, analgesics, diuretics, rehydration and physical chemotherapy are included, as well as the infected wounds that can be managed at the bedside. (2) Grade II: Complications requiring medications in addition to those listed in Class I. Blood transfusion and total parenteral nutrition are also included. (3) Grade III: Complications requiring surgical intervention, intervention, endoscopic treatment, and total parenteral nutrition. Those require general anesthesia are categorized into level IIIa, and those do not into level IIIb. (4) Level IV: Life-threatening complications (including central nervous system complications) that require intensive care unit treatment, with single-organ failure at level IVa (including the need for dialysis) and multi-organ failure at level IVb. (5) Grade V: death.

## Statistical analysis

Normally distributed measurement data were represented by mean and standard deviation, with difference compared by Student's *t*-tests. Non-normally distributed continuous variables were reported as the median with interquartile range and were compared by Mann–Whitney *U*-tests, with categorical variables compared by $X^2$ test or Fisher's exact test. Univariate analysis covered all potential indicators, including preoperative, intraoperative and postoperative patient-related factors. Multivariate logistic regression analysis including the potential factors with $P \leq 0.05$ in univariate analysis was conducted to identify the risk factors associated with all and major complications after LPD. Potential interactions between these factors and the level of complications were also examined. Results were represented by *P*-values, odd ratios(ORs)and 95% confidence intervals (CIs). *P*-value of ≤0.05 was considered statistically significant difference. All statistic analyses were performed using software SPSS Version 25.0.

## Results

### Cohort basic characteristics

From April 2015 to November 2021,824 patients underwent LPD at the First Affiliated Hospital of JiLin University. 31 patients were excluded owing to the lack of data. The basic characteristic and surgical details of the patients were listed in Table 1. The median age of 793 patients was 60 (IQR: 52–66) years, composed of 442 (55.7%) males and 351 (44.3%) females.

TABLE 1 Baseline characteristics of the study cohort.

| Variables | Value |
|---|---|
| Total | 793 (100) |
| Sex (female/male, *n*%) | 351 (44.3)/442 (55.7) |
| Age (years, IQR) | 60 (52–66) |
| BMI (kg/m$^2$, SD) | 22.97 ± 3.23 |
| Preoperative CA19-9 (U/I, SD) | 154.05 ± 197.59 |
| Preoperative TBIL (mmol/L,SD) | 94.23 ± 85.67 |
| Hypertension (yes/no, *n*%) | 112 (14.1)/681 (85.9) |
| Diabetes (yes/no, *n*%) | 103 (13.0)/690 (87.0) |
| Virus hepatitis (yes/no, *n*%) | 25 (3.2)/768 (96.8) |
| Preoperative biliary drainage (yes/no, *n*%) | 321 (40.5)/472 (59.5) |
| ASA grade (I/II/III, *n*%) | 34 (4.3)/640 (80.7)/119 (15.0) |
| History of abdominal surgery (yes/no, *n*%) | 139 (17.5)/654 (82.5) |
| Vascular variation (yes/no, *n*%) | 560 (70.6)/233 (29.4) |
| Introperative bleeding (ml, IQR) | 50 (20–100) |
| Intraoperative transfusions (yes/no, *n*%) | 145 (18.3)/648 (81.7) |
| Operation time (min, SD) | 191.02 ± 66.90 |
| Pancreas texture (firm/middle/soft, *n*%) | 234 (29.5)/135 (17.0)/424 (53.5) |
| Size of pancreatic duct (>3/≤3, *n*%) | 385 (48.5)/408 (51.5) |

BMI, body mass index; IQR, interquartile range; SD, mean; CA19-9, cancer antigen 19-9, TBIL, total bilirubin; ASA, American Society of Anesthesiologists.

Mean total bilirubin was 94.23 ± 85.67 mmol/L. Patients with mean cancer antengin19-9 of 154.05 ± 197.59 U/I.321 (40.5%) underwent ultrasonic-guided bile drainage due to hyperbilirubinemia before surgery.112 (14.1%) had hypertension, 103 (13.0%) had diabetes and 25 (3.2%) had virus hepatitis. 34 (4.3%) patients were classified as ASA I, 640 (80.7%) as ASA II and 119 (15.0%) as ASA III. Median blood loss was 50 (20–100) ml. Mean operation time was 191.02 ± 66.90 min. Pancreatic specimens were soft in 424 (53.5%) patients, middle in 135 (17.0%) patients and firm in 234 (29.5%) patients. 408 (51.5%) patients was found to exhibit pancreatic duct diameter ≤ 3 mm 385 (48.5%) patients found >3 mm. Other baseline characteristics, intraoperative details are described in Table 1.

LPPC occurred in 260 (32.8%) patients, with 169 (21.3%) patients developing POPF, 44 (5.5%)patients developing biliary fistula, 17 (2.1%) patients developing DGE, 55 (6.9%) patients developing PPH, 7 (0.8%) patients developing intestinal fistula, 59 (7.4%) patients developing abdominal infections and 28 (3.5%) patients developing pulmonary complication. According to CDC, the LPPC of all patients could be divided into five grades (Grade I–V), of which grade III was subdivided into grade IIIa and grade IIIb according to whether there was invasive operation under general anesthesia. POPF was determined to be the most common complication after LPD in our study. In Table 2 the detailed classification of complications is shown. The number of patients with C–D grade I, II, IIIa, IIIb, IV and V was 83 (31.9%), 91 (35.0%), 38 (14.6%), 24 (9.2%), 9 (3.5%) and 15 (5.8%). The grade I–II was classified as mild LPPC and grade III–V as severe LPPC. 174 (66.9%) patients were categorized with grade I–II and 86 (33.1%) with grade III–V. The 793 patients were further divided into two groups: 707 (89.2%) patients with no or mild LPPC, and 86 (10.8%) patients with severe LPPC, among which 15 (1.9%) patients experienced postoperative death, 6 (40.0%) died of multiple organ failure due to severe

TABLE 2 Clavien–Dindo classification of postoperative complications.

| Complications | Total | Grade I | Grade II | Grade IIIa | Grade IIIb | Grade IV | Grade V |
|---|---|---|---|---|---|---|---|
| Pancreatic fistula | 169 | 60 | 61 | 30 | 3 | 6 | 9 |
| Hemorrhage | 55 | 1 | 2 | 8 | 24 | 2 | 7 |
| Delayed gastric emptying | 17 | 1 | 8 | 3 | 1 | 4 | 0 |
| Billary fistula | 44 | 13 | 12 | 14 | 2 | 1 | 2 |
| Abdominal infections | 59 | 6 | 15 | 18 | 7 | 7 | 6 |
| Pulmonary complication | 28 | 8 | 7 | 5 | 1 | 2 | 5 |
| Intestinal fistula | 7 | 0 | 1 | 3 | 1 | 1 | 1 |

postoperative infection, 4 (26.6%) died due to abdominal bleeding and failure in stopping bleeding after secondary laparotomy, 3 (20.0%) died of respiratory failure, 1 (6.7%) died two weeks after discharge with a large amount of blood visible in the abdominal drainage tube, which was considered to be arterial stump bleeding, and 1 (6.7%) died of pulmonary embolism.

## Risk factors of LPPC

The results of univariate analysis of postoperative complications and severe complications were listed in Table 3 and those of multivariate analysis in Table 4. In univariate analysis, gender ($P = 0.006$), soft pancreatic texture ($P < 0.001$) and pancreatic duct diameter $\leq 3$ mm ($P = 0.009$) were significantly associated with LPPC, while BMI ($P = 0.027$), preoperative total bilirubin ($P = 0.010$), preoperative biliary drainage ($P = 0.049$) and intraoperative blood transfusion ($P = 0.015$) were associated with LPPC. Severe LPPC was significantly associated with preoperative TBIL > 170 mmol/L ($P < 0.001$) and intraoperative blood transfusion ($P = 0.002$), appearing to be related with size of pancreatic duct ($P = 0.045$).

In multivariate Logistic regression analysis, pancreatic texture ($P < 0.001$, OR = 1.399, 95% CI: 1.170–1.673), intraoperative blood transfusion ($P = 0.033$, OR = 1.517, 95% CI, 1.034–2.226), gender ($P = 0.038$, OR = 1.396, 95% CI: 1.019–1.911) and preoperative TBIL > 170 mmol/L ($P = 0.029$, OR = 1.523, 95% CI: 1.043–2.224) were independent risk factors for postoperative complications of LPD. Severe LPPC was revealed to be independently associated with preoperative TBIL > 170 mmol/L ($P = 0.001$, OR = 2.313, 95% CI: 1.406–3.807) and intraoperative transfusion ($P = 0.004$, OR = 2.123, 95% CI: 1.278–3.529).

Analysis of differences between mild and severe complications.

As shown in Table 5, in comparison with mild complications (grade I–II), severe complications (grade III–V) were associated with preoperative CA19-9 ($P = 0.019$), preoperative TBIL > 170 mmol/L ($P = 0.015$), and total operation time ($P = 0.036$). Multivariate analysis suggested preoperative TBIL > 170 mmol/L ($P = 0.036$, OR = 1.901, 95% CI: 1.043–3.484) as an independent risk factor.

The hospital stay of patients with complications of all grades was evaluated as grade I (19.06 ± 4.575), II (26.82 ± 6.251), IIIa (38.66 ± 9.737), IIIb (30.33 ± 12.815), IV (72.78 ± 10.721) and V (22.60 ± 11.564). As depicted in Figure 1, except for patients

TABLE 3 Univariate analysis of postoperative complications and severe complications.

| | | No-LPPC $n = 533$ | LPPC $n = 260$ | *P*-value | Grade 0–II $n = 707$ | Grade III–V $n = 86$ | *P*-value |
|---|---|---|---|---|---|---|---|
| Age | <65/≥65 | 365/168 | 178/82 | 0.996 | 490/217 | 53/33 | 0.148 |
| Sex | Female/male | 254/279 | 97/163 | 0.006** | 315/392 | 36/50 | 0.635 |
| BMI (kg/m$^2$) | ≤23.9/>23.9 | 366/167 | 158/102 | 0.027* | 471/236 | 53/33 | 0.356 |
| CA19-9 (U/I) | ≤100/>100 | 302/231 | 165/95 | 0.068 | 421/286 | 46/40 | 0.281 |
| Vascular variation | yes/no | 153/380 | 80/180 | 0.549 | 209/498 | 24/62 | 0.750 |
| Preoperative TBIL (mmol/L) | ≤170/>170 | 448/85 | 199/61 | 0.010** | 589/118 | 58/28 | 0.000** |
| Preoperative biliary drainage | Yes/no | 203/330 | 118/142 | 0.049* | 279/428 | 42/44 | 0.094 |
| Hypertension | Yes/no | 77/456 | 35/225 | 0.708 | 100/607 | 12/74 | 0.962 |
| Diabetes | Yes/no | 65/468 | 38/222 | 0.341 | 91/618 | 12/74 | 0.778 |
| Virus hepatitis | Yes/no | 13/520 | 12/248 | 0.100 | 22/685 | 3/83 | 0.850 |
| History of abdominal surgery | Yes/no | 92/441 | 47/213 | 0.777 | 121/586 | 18/68 | 0.380 |
| ASA grade | ≤II/>II | 454/79 | 220/40 | 0.835 | 601/106 | 73/13 | 0.976 |
| Operation time (min) | ≤300/>300 | 492/41 | 242/18 | 0.698 | 658/49 | 76/10 | 0.117 |
| Pancreas texture | Firm/middle/soft | 174/100/259 | 60/35/165 | 0.000* | 373/120/214 | 51/15/20 | 0.386 |
| Size of pancreatic duct | >3/≤3 | 276/257 | 109/151 | 0.009** | 352/355 | 33/53 | 0.045* |
| Introperative bleeding (ml) | ≤400/>400 | 506/27 | 241/19 | 0.205 | 668/39 | 79/7 | 0.326 |
| Intraoperative transfusions | Yes/no | 85/448 | 60/200 | 0.015* | 119/588 | 26/60 | 0.002** |

BMI, body mass index; CA19-9, cancer antigen 19-9, TBIL, total bilirubin; ASA, American Society of Anesthesiologists. Grade 0–II, no complications and Clavien–Dindo classification grade I–II.

*$P < 0.05$.

**$P < 0.01$.

TABLE 4 Multivariate analysis of postoperative complications and severe complications.

| | | OR-value | 95% confidence interval | | *P*-value |
|---|---|---|---|---|---|
| | | | Lower | Upper | |
| **No-LPPC/LPPC** | | | | | |
| Pancreas texture | Firm/Middle/Soft | 1.399 | 1.170 | 1.673 | 0.000 |
| Intraoperative transfusions | no | 1 | | | |
| | yes | 1.517 | 1.034 | 2.226 | 0.033 |
| sex | Female | 1 | | | |
| | Male | 1.396 | 1.019 | 1.911 | 0.038 |
| Preoperative TBIL (mmol/L) | ≤170 | 1 | | | |
| | >170 | 1.523 | 1.043 | 2.224 | 0.029 |
| **Grade 0–II/III–V** | | | | | |
| Preoperative TBIL (mmol/L) | ≤170 | 1 | | | |
| | >170 | 2.313 | 1.406 | 3.807 | 0.001 |
| Intraoperative transfusions | 否 | 1 | | | |
| | 是 | 2.123 | 1.278 | 3.529 | 0.004 |

LPPC, post-laparoscopic pancreaticoduodenectomy complications; TBIL, total bilirubin; Grade 0–II, no complications and Clavien–Dindo classification grade I–II.

who died, the length of postoperative hospital stay was generally prolonged with the elevation of the LPPC grade.

## Discussion

PD is the primary choice in the treatment of periampullary tumor, which has even become a representative of advanced surgery celebrated by its high degree of difficulty. In recent years, laparoscopy has been favored by surgeons with its advantages of small trauma, low pain and quick recovery, as minimally invasive Whipple has been widely carried out in general surgery around the world (17–21). The service of this operation involves a number of organs and makes great impacts in human anatomy. Despite the modification of technique and clinical nursing level in recent years, its complication rate is still reported as high as 50%–60% (22, 23). In this study, the complications rate was reported to be only 32.8% (260/793). There has always existed a competitive relationship between the global general surgery and the medical center in completing LPD with a low postoperative mortality rate, which will undoubtedly win a better public praise and reputation, leading the forefront of surgery at home and abroad. Therefore, the existence of a unified standard to compare the efficacy of LPD in different regions and countries is required, and the C–D classification provides such a tool, which as a grading standard has been referred to in many surgical fields (24–26).

The C–D grading itself has several advantages (11). First, distinguished from the traditional single-system study, it evaluates the surgical efficacy from the overall multi-system. Secondly, it can prepare for the assessment of potential independent risk factors for surgery-related complications. Finally, it can contribute to exploring the factors that may aggravate the complications, thus fundamentally reducing the occurrence of such events, which benefits the surgical field as a whole.

This study concluded that soft pancreas could serve as an independent risk factor for postoperative complications of LPD, which is also consistent with the view of most scholars. The soft pancreas (26, 27) generally has a good exocrine function with the capability to secrete a large amount of pancreatic fluid. During

TABLE 5 Univariate and multivariate analysis of complication grading.

| | | Univariate | | | Multivariate | | | |
|---|---|---|---|---|---|---|---|---|
| | | Grade I–II *n* = 174 | Grade III–V *n* = 86 | *P*-value | OR-value | 95%CI | | *P*-value |
| | | | | | | Lower | upper | |
| Age | <65/≥65 | 125/49 | 53/33 | 0.095 | | | | |
| Sex | Female/male | 61/113 | 36/50 | 0.286 | | | | |
| BMI (kg/m$^2$) | ≤23.9/>23.9 | 105/69 | 53/33 | 0.842 | | | | |
| CA19-9 (U/I) | ≤100/>100 | 119/55 | 46/40 | 0.019* | | | | |
| Vascular variation | Yes/no | 56/118 | 24/62 | 0.482 | | | | |
| Preoperative TBIL (mmol/L) | ≤170/>170 | 141/33 | 58/28 | 0.015* | 1.906 | 1.043 | 3.484 | 0.036* |
| Preoperative biliary drainage | Yes/no | 76/98 | 42/44 | 0.432 | | | | |
| Hypertension | Yes/no | 23/151 | 12/74 | 0.870 | | | | |
| Diabetes | Yes/no | 26/148 | 12/74 | 0.832 | | | | |
| Virus hepatitis | Yes/no | 9/165 | 3/83 | 0.543 | | | | |
| History of abdominal surgery | Yes/no | 29/145 | 18/68 | 0.401 | | | | |
| ASA grade | ≤II/>II | 147/27 | 73/13 | 0.933 | | | | |
| Operation time (min) | ≤300/>300 | 166/8 | 76/10 | 0.036* | | | | |
| Pancreas texture | Firm/middle/soft | 40/20/114 | 20/15/51 | 0.394 | | | | |
| Size of pancreatic duct | >3/≤3 | 76/98 | 33/53 | 0.415 | | | | |
| Introperative bleeding (ml) | ≤400/>400 | 162/12 | 79/7 | 0.717 | | | | |
| Intraoperative transfusions | Yes/no | 34/140 | 26/60 | 0.054 | | | | |

BMI, body mass index;CA19-9,cancer antigen 19-9,TBIL,total bilirubin;ASA, American Society of Anesthesiologists.
*$P < 0.05$.

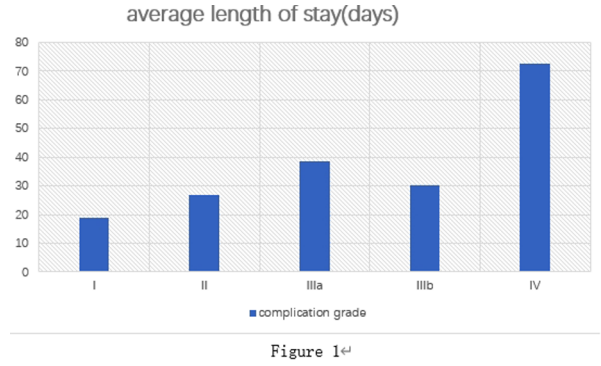


FIGURE 1
Length of stay for complications with different C-D grades.

pancreatic jejunal anastomosis after LPD, it is easy to corrode the anastomotic vessels and tissues. Secondly, when the soft pancreas is anastomosed with the residual pancreas, cutting effect is more likely appear by the suture and lead to pancreatic damage, which will cause POPF, resulting in bleeding, abdominal infection, sepsis and other complications, which has reached a consensus in academic (27–30). The pancreas with low density is more sensitive to inflammation compared to those with high fibrosis. With the subside of inflammation after surgery, the volume of the remaining pancreas will be slightly reduced, and the gap between the suture and the tissue will also develop, which also provides an opportunity for pancreas fluid leakage.

The diameter of pancreatic duct (5, 28, 29) is related to LPPC, which with excessively thin duct is associated to the higher occurrence of damage in the pancreas when anastomosing with jejunum mucosa, and difficult to exact anastomosis. Another study in our center (27) demonstrated the diameter of small pancreatic duct as an independent risk factor for postoperative POPF (OR: 30.277, 95% CI: 10.578–86.655, $P < 0.001$), which was also verified in other studies. However, the expanded sample size resulted in the statistically insignificant diameter of pancreatic duct in the multivariate analysis in the present study. We speculate the other LPPC resulting from pancreatic juice when POPF occurs after LPD, such as PPH, abdominal infection, etc., so the diameter of pancreatic duct is considered to be related to LPPC. However, due to the absence of uniform standard for the measurement of the diameter, which is thus estimated roughly according to the experience of the operator, these data may be biased, further verification from other medical centers is required.

In the study, male sex was a risk factor of LPPC, but exhibited no significant association. Most studies (31–33) have not reported that gender differences affect the rate of postoperative complications. We considered this result to be related to the living habits of people in northeast China. In northeastern China, table culture is a weighted means of communication, especially alcohol consumption, which is a main cause of chronic pancreatitis. Although the hard pancreas are almost universally accepted more likely to reduce the incidence of postoperative complications in terms of technique, some scholars (34, 35) argue that the excessive fibrosis of the pancreas can affect the development of pancreatic anastomosis stoma, tending to leave gaps between the pancreas and jejunum in the process of stitching, and possible lacuna between pancreatic duct and supporting tube, which will be the hidden trouble to the patient outcome. It is also believed that men and women have different fat distribution and patients with more abdominal fat also have more fat in the pancreas, which affects the texture of the pancreas (36–38) and produce a certain impact on prognosis. However, the effect of gender (39–41) or history of chronic pancreatic on LPD prognosis still requires further study due to lack of enrolled studies, which may have a strong regional character.

Despite the necessity of perioperative blood transfusion for patients with large blood loss during major surgery, it has been determined that blood transfusion is significantly associated with postoperative complications (42). We found that intraoperative blood transfusion was an independent risk factor for LPPC, possibly related to the systemic inflammatory response that blood transfusion may elicit after surgery. Large transfusions of red blood cells can also result in dilution clotting factors deficiency (43–46). Dirk J et al. (47) reported that the odds ratio for exposure to intraoperative blood transfusion in patients was 1.74. Some scholars (48) have concluded a linear correlation between blood transfusion and postoperative morbidity. The elevated risk of postoperative infection may be resulted from the immunosuppression caused by blood transfusion, which inhibits the activity of immune cells, such as T-cells and nature killer cells, and may promote the release of some growth factors, thus inducing tumor recurrence. Therefore, the indication of blood transfusion should be strictly grasped.

High bilirubin itself is a manifestation of liver damage. In surgery, cholestatic liver damage is often caused by biliary obstruction, which results in insufficient synthesis of coagulation factors and increased risk of postoperative bleeding (6, 49). There also have studies clearly reporting a higher incidence of liver failure or multiple organ failure in patients with high preoperative bilirubin levels (50–53). Vitamin K deficiency is common in patients undergoing preoperative bile drainage, which affects clotting factors synthesis, as well as in patients with obstructive liver injury. It has been suggested that mildly elevated bilirubin induced platelet activation *via* mechanism related to collagen-induced platelet activation, thereby inhibiting coagulation (54). All of these increase the risk of bleeding after surgery. This study suggests hyperbilirubinemia as an independent risk factor for postoperative complications of LPD, and is closely associated with the incidence of severe complications, which may even contribute to postoperative deterioration of the disease. One study (27) from a large capacity center in western China covering 1056 patients also identified hyperbilirubinemia as an independent risk factor for LPPC, especially highly correlated with Grade V ($P = 0.042$, 95% CI: 1.849 to 4.789, OR = 2.017). In univariate analysis, preoperative biliary drainage exhibited no statistical significance after excluding the interference of other factors after inclusion in regression model. Some scholars (55–57) believe that preoperative biliary drainage aggravates the risk of postoperative biliary tract infection, while it is undeniable that the alleviation of jaundice by preoperative drainage can significantly improve liver function with

the potential to optimize the prognosis of patients (58). In multivariate analysis of this study, the *P*-values of bilirubin were 0.029, 0.001 and 0.036, respectively. There showed statistically significance between mild and severe complications ($P = 0.001$). Consideration of bilirubin not only increases the incidence of LPPC, but also may lead to the development of severe complications.

As shown in **Figure 1**, the hospital stay after surgery is generally extended with the improvement of LPPC level. Therefore, the C–D grading system is expected to improve perioperative patient management, shorten hospital stay, reduce medical costs and patient economic pressure.

In summary, we concluded the significant association of the results of CDC with risk factors for LPPC, which may accurately predict the major complications. This grading system could provide a reliable means of quality assessment in surgical procedures and contribute to date comparison among different medical bases and therapies. It may also be widely applied in abdominal surgery in the future. All of this will help modify the quality of minimally invasive surgery, contributing shorter hospital stay and decreased financial costs.

According to this study, we believe that the C–D system in clinical management can predict the postoperative recovery of patients. By analyzing the differences between complications of different severity, we found that certain factors such as hyperbilirubinemia and intraoperative blood transfusion were statistically significant, which suggests that we should pay more attention to the presence of such factors in patient management and try to correct preoperative hyperbilirubin as much as possible. We hope to establish a new scoring system. We can score by relevant preoperative risk factors, and then estimate the possibility of complications at all levels after surgery. However, the sample size of our center is limited, and we are unable to complete it for the time being. In addition, we are collecting new data. When the sample size is sufficient, we will further verify the results of this study and establish a new scoring system as far as possible. It is also hoped that other large capacity centers at home and abroad can further verify this experiment.

The study, as a single-center retrospective analysis, also has some limitations. First, the data were collected prospectively, possibly biasing in the process of information collection, and selection bias may exist in the selection of research objects. Second, the sample size is only concentrated in one region. Third, some variables were not considered in the study due to different treatment concepts. The results of this study require to be further verified by multi-center, accurately designed and reliable prospective studies in large-capacity centers to obtain more valuable results.

## Data availability statement

The original contributions presented in the study are included in the article/Supplementary Material, further inquiries can be directed to the corresponding author.

## Author contributions

YHL: project development; XYS: manuscript writing; YM: data analysis; HYS: data collection. All authors contributed to the article and approved the submitted version.

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## Conflict of interest

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

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## References

1. Liu M, Ji S, Xu W, Liu W, Qin Y, Hu Q, et al. Laparoscopic pancreaticoduodenectomy: are the best times coming? *World J Surg Oncol.* (2019) 17(1):81. doi: 10.1186/s12957-019-1624-6

2. Espin Alvarez F, García Domingo MI, Cremades Pérez M, Herrero Fonollosa E, Navinés López J, Camps Lasa J, et al. Highs and lows in laparoscopic pancreaticoduodenectomy. *Cir Esp.* (2021) 99(8):593–601. doi: 10.1016/j.cireng.2021.08.001

3. Wang M, Peng B, Liu J, Yin X, Tan Z, Liu R, et al. Practice patterns and perioperative outcomes of laparoscopic pancreaticoduodenectomy in China: a retrospective multicenter analysis of 1029 patients. *Ann Surg.* (2021) 273(1):145–53. doi: 10.1097/SLA.0000000000003190

4. Sandini M, Ruscic KJ, Ferrone CR, Qadan M, Eikermann M, Warshaw AL, et al. Major complications independently increase long-term mortality after pancreatoduodenectomy for cancer. *J Gastrointest Surg.* (2019) 23(10):1984–90. doi: 10.1007/s11605-018-3939-y

5. Braga M, Capretti G, Pecorelli N, Balzano G, Doglioni C, Ariotti R, et al. A prognostic score to predict Major complications after pancreaticoduodenectomy. *Ann Surg.* (2011) 254(5):702–7; discussion 707–8. doi: 10.1097/SLA.0b013e31823598fb

6. Das S, Ray S, Mangla V, Mehrotra S, Lalwani S, Mehta N, et al. Post pancreaticoduodenectomy hemorrhage: a retrospective analysis of incidence, risk factors and outcome. *Saudi J Gastroenterol.* (2020) 26(6):337. doi: 10.4103/sjg.SJG_145_20

7. Kitahata Y, Hirono S, Kawai M, Okada K-I, Miyazawa M, Shimizu A, et al. Intensive perioperative rehabilitation improves surgical outcomes after pancreaticoduodenectomy. *Langenbecks Arch Surg.* (2018) 403(6):711–8. doi: 10.1007/s00423-018-1710-1

8. Wente MN, Veit JA, Bassi C, Dervenis C, Fingerhut A, Gouma DJ, et al. Postpancreatectomy hemorrhage (PPH)–an international study group of pancreatic surgery (ISGPS) definition. *Surgery.* (2007) 142(1):20–5. doi: 10.1016/j.surg.2007.02.001

9. Bassi C, Dervenis C, Butturini G, Fingerhut A, Yeo C, Izbicki J, et al. Postoperative pancreatic Fistula: an international study group (ISGPF) definition. *Surgery.* (2005) 138(1):8–13. doi: 10.1016/j.surg.2005.05.001

10. Yeo CJ, Barry MK, Sauter PK, Sostre S, Lillemoe KD, Pitt HA, et al. Erythromycin accelerates gastric emptying after pancreaticoduodenectomy. A prospective, randomized, placebo-controlled trial. *Ann Surg.* (1993) 218(3):229–37; discussion 237–238. doi: 10.1097/00000658-199309000-00002

11. Dindo D, Demartines N, Clavien P-A. Classification of surgical complications: a new proposal with evaluation in a cohort of 6336 patients and results of a survey. *Ann Surg.* (2004) 240(2):205–13. doi: 10.1097/01.sla.0000133083.54934.ae

12. Katayama H, Kurokawa Y, Nakamura K, Ito H, Kanemitsu Y, Masuda N, et al. Extended clavien–dindo classification of surgical complications: japan clinical oncology group postoperative complications criteria. *Surg Today.* (2016) 46(6):668–85. doi: 10.1007/s00595-015-1236-x

13. Fransen LFC, Berkelmans GHK, Asti E, van Berge Henegouwen MI, Berlth F, Bonavina L, et al. The effect of postoperative complications after minimally invasive esophagectomy on long-term survival: an international multicenter cohort study. *Ann Surg.* (2021) 274(6):e1129–37. doi: 10.1097/SLA.0000000000003772

14. Lee D-K, Frye A, Louis M, Koshy AN, Tosif S, Yii M, et al. Postoperative complications and hospital costs following small bowel resection surgery. *PloS One.* (2020) 15(10):e0241020. doi: 10.1371/journal.pone.0241020

15. Wente MN, Bassi C, Dervenis C, Fingerhut A, Gouma DJ, Izbicki JR, et al. Delayed gastric emptying (DGE) after pancreatic surgery: a suggested definition by the international study group of pancreatic surgery (ISGPS). *Surgery.* (2007) 142(5):761–8. doi: 10.1016/j.surg.2007.05.005

16. Koch M, Garden OJ, Padbury R, Rahbari NN, Adam R, Capussotti L, et al. Bile leakage after hepatobiliary and pancreatic surgery: a definition and grading of severity by the international study group of liver surgery. *Surgery.* (2011) 149(5):680–8. doi: 10.1016/j.surg.2010.12.002

17. Zhang H, Lan X, Peng B, Li B. Is total laparoscopic pancreaticoduodenectomy superior to open procedure? A meta-analysis. *World J Gastroenterol.* (2019) 25(37):5711–31. doi: 10.3748/wjg.v25.i37.5711

18. Shin SH, Kim Y-J, Song KB, Kim S-R, Hwang DW, Lee JH, et al. Totally laparoscopic or robot-assisted pancreaticoduodenectomy versus open surgery for periampullary neoplasms: separate systematic reviews and meta-analyses. *Surg Endosc.* (2017) 31(9):3459–74. doi: 10.1007/s00464-016-5395-7

19. Wang X, Cai Y, Jiang J, Peng B. Laparoscopic pancreaticoduodenectomy: outcomes and experience of 550 patients in a single institution. *Ann Surg Oncol.* (2020) 27(11):4562–73. doi: 10.1245/s10434-020-08533-3

20. Nickel F, Haney CM, Kowalewski KF, Probst P, Limen EF, Kalkum E, et al. Laparoscopic versus open pancreaticoduodenectomy: a systematic review and meta-analysis of randomized controlled trials. *Ann Surg.* (2020) 271(1):54–66. doi: 10.1097/SLA.0000000000003309

21. Wang M, Li D, Chen R, Huang X, Li J, Liu Y, et al. Minimally invasive treatment group in the pancreatic disease branch of China's international exchange and promotion association for medicine and healthcare (MITG-P-CPAM). laparoscopic versus open pancreatoduodenectomy for pancreatic or periampullary tumours: a multicentre, open-label, randomised controlled trial. *Lancet Gastroenterol Hepatol.* (2021) 6(6):438–47. doi: 10.1016/S2468-1253(21)00054-6

22. van der Gaag NA, Harmsen K, Eshuis WJ, Busch ORC, van Gulik TM, Gouma DJ. Pancreatoduodenectomy associated complications influence cancer recurrence and time interval to death. *Eur J Surg Oncol J Eur Soc Surg Oncol Br Assoc Surg Oncol.* (2014) 40(5):551–8. doi: 10.1016/j.ejso.2013.12.012

23. Kawaida H, Kono H, Hosomura N, Amemiya H, Itakura J, Fujii H, et al. Surgical techniques and postoperative management to prevent postoperative pancreatic Fistula after pancreatic surgery. *World J Gastroenterol.* (2019) 25(28):3722–37. doi: 10.3748/wjg.v25.i28.3722

24. Téoule P, Bartel F, Birgin E, Rückert F, Wilhelm TJ. The clavien–dindo classification in pancreatic surgery: a clinical and economic validation. *J Invest Surg.* (2019) 32(4):314–20. doi: 10.1080/08941939.2017.1420837

25. Clavien PA, Barkun J, de Oliveira ML, Vauthey JN, Dindo D, Schulick RD, et al. The clavien–dindo classification of surgical complications: five-year experience. *Ann Surg.* (2009) 250(2):187–96. doi: 10.1097/SLA.0b013e3181b13ca2

26. Wang W, Babu S, Wang L, Chen Y, Tian B, He H. Use of clavien–dindo classification in evaluating complications following pancreaticoduodenectomy in 1,056 cases: a retrospective analysis from one single institution. *Oncol Lett.* (2018) 16(2):2023–9. doi: 10.3892/ol.2018.8798

27. Niu C, Chen Q, Liu S, Zhang W, Jiang P, Liu Y. Clinical validation of the risk scoring systems of postoperative pancreatic Fistula after laparoscopic pancreatoduodenectomy in Chinese cohorts: a single-center retrospective study. *Surgery.* (2022) 171(4):1051–7. doi: 10.1016/j.surg.2021.08.013

28. Mungroop TH, van Rijssen LB, van Klaveren D, Smits FJ, van Woerden V, Linnemann RJ, et al. Dutch Pancreatic cancer group. Alternative Fistula risk score for pancreatoduodenectomy (a-FRS): design and international external validation. *Ann Surg.* (2019) 269(5):937–43. doi: 10.1097/SLA.0000000000002620

29. Ryu Y, Shin SH, Park DJ, Kim N, Heo JS, Choi DW, et al. Validation of original and alternative Fistula risk scores in postoperative pancreatic Fistula. *J Hepato-Biliary-Pancreat Sci.* (2019) 26(8):354–9. doi: 10.1002/jhbp.638

30. Hong SS, Chong JU, Hwang HK, Lee WJ, Kang CM. Laparoscopic pancreaticoduodenectomy reduces incidence of clinically relevant postoperative pancreatic Fistula in soft pancreas with a smaller than 2 mm pancreatic duct. *Surg Endosc.* (2021) 35(12):7094–103. doi: 10.1007/s00464-020-08226-8

31. Gaujoux S, Cortes A, Couvelard A, Noullet S, Clavel L, Rebours V, et al. Fatty pancreas and increased body mass Index are risk factors of pancreatic Fistula after pancreaticoduodenectomy. *Surgery.* (2010) 148(1):15–23. doi: 10.1016/j.surg.2009.12.005

32. Vining CC, Kuchta K, Schuitevoerder D, Paterakos P, Berger Y, Roggin KK, et al. Risk factors for complications in patients undergoing pancreaticoduodenectomy: a NSQIP analysis with propensity score matching. *J Surg Oncol.* (2020) 122(2):183–94. doi: 10.1002/jso.25942

33. Lof S, Vissers FL, Klompmaker S, Berti S, Boggi U, Coratti A, et al. European Consortium on minimally invasive pancreatic surgery (E-MIPS). risk of conversion to open surgery during robotic and laparoscopic pancreatoduodenectomy and effect on outcomes: international propensity score-matched comparison study. *Br J Surg.* (2021) 108(1):80–7. doi: 10.1093/bjs/znaa026

34. Senthilnathan P, Subrahmaneswara Babu N, Vikram A, Sabnis SC, Srivatsan Gurumurthy S, Anand Vijai N, et al. Laparoscopic longitudinal pancreatojejunostomy and modified frey's operation for chronic calcific pancreatitis. *BJS Open.* (2019) 3(5):666–71. doi: 10.1002/bjs5.50185

35. Martin RF, Marion MD. Resectional therapy for chronic pancreatitis. *Surg Clin North Am.* (2007) 87(6):1461–75; ix. doi: 10.1016/j.suc.2007.09.006

36. Abe T, Amano H, Kobayashi T, Hanada K, Hattori M, Nakahara M, et al. Preoperative anthropomorphic and nutritious Status and Fistula risk score for predicting clinically relevant postoperative pancreatic Fistula after pancreaticoduodenectomy. *BMC Gastroenterol.* (2020) 20(1):264. doi: 10.1186/s12876-020-01397-7

37. House MG, Fong Y, Arnaoutakis DJ, Sharma R, Winston CB, Protic M, et al. Preoperative predictors for complications after pancreaticoduodenectomy: impact of BMI and body fat distribution. *J Gastrointest Surg.* (2008) 12(2):270–8. doi: 10.1007/s11605-007-0421-7

38. Ecker BL, McMillan MT, Allegrini V, Bassi C, Beane JD, Beckman RM, et al. Risk factors and mitigation strategies for pancreatic Fistula after distal pancreatectomy: analysis of 2026 resections from the international, multi-institutional distal pancreatectomy study group. *Ann Surg.* (2019) 269(1):143–9. doi: 10.1097/SLA.0000000000002491

39. Hu B-Y, Wan T, Zhang W-Z, Dong J-H. Risk factors for postoperative pancreatic Fistula: analysis of 539 successive cases of pancreaticoduodenectomy. *World J Gastroenterol.* (2016) 22(34):7797–805. doi: 10.3748/wjg.v22.i34.7797

40. Ellis RJ, Gupta AR, Hewitt DB, Merkow RP, Cohen ME, Ko CY, et al. Risk factors for post-pancreaticoduodenectomy delayed gastric emptying in the absence of pancreatic Fistula or intra-abdominal infection. *J Surg Oncol.* (2019) 119(7):925–31. doi: 10.1002/jso.25398

41. Mungroop TH, Klompmaker S, Wellner UF, Steyerberg EW, Coratti A, D'Hondt M, et al. European Consortium on minimally invasive pancreatic surgery (E-MIPS). updated alternative Fistula risk score (ua-FRS) to include minimally invasive pancreatoduodenectomy: pan-European validation. *Ann Surg.* (2021) 273(2):334–40. doi: 10.1097/SLA.0000000000003234

42. Polanco PM, Zenati MS, Hogg ME, Shakir M, Boone BA, Bartlett DL, et al. An analysis of risk factors for pancreatic Fistula after robotic pancreaticoduodenectomy: outcomes from a consecutive series of standardized pancreatic reconstructions. *Surg Endosc.* (2016) 30(4):1523–9. doi: 10.1007/s00464-015-4366-8

43. Zhang L, Liao Q, Zhang T, Dai M, Zhao Y. Blood transfusion is an independent risk factor for postoperative serious infectious complications after pancreaticoduodenectomy. *World J Surg.* (2016) 40(10):2507–12. doi: 10.1007/s00268-016-3553-7

44. Yang J-C, Sun Y, Xu C-X, Dang Q-L, Li L, Xu Y-G, et al. Coagulation defects associated with massive blood transfusion: a large multicenter study. *Mol Med Rep.* (2015) 12(3):4179–86. doi: 10.3892/mmr.2015.3971

45. Sihler KC, Napolitano LM. Complications of massive transfusion. *Chest.* (2010) 137(1):209–20. doi: 10.1378/chest.09-0252

46. Peng Y-P, Zhu X-L, Yin L-D, Zhu Y, Wei J-S, Wu J-L, et al. Risk factors of postoperative pancreatic Fistula in patients after distal pancreatectomy: a systematic review and meta-analysis. *Sci Rep.* (2017) 7(1):185. doi: 10.1038/s41598-017-00311-8

47. Gouma DJ, van Geenen RCI, van Gulik TM, de Haan RJ, de Wit LT, Busch ORC, et al. Rates of complications and death after pancreaticoduodenectomy: risk factors and the impact of hospital volume. *Ann Surg*. (2000) 232(6):786–95. doi: 10.1097/00000658-200012000-00007

48. Woods BI, Rosario BL, Chen A, Waters JH, Donaldson W, Kang J, et al. The association between perioperative allogeneic transfusion volume and postoperative infection in patients following lumbar spine surgery. *J Bone Joint Surg Am*. (2013) 95(23):2105–10. doi: 10.2106/JBJS.L.00979

49. Lu J, Ding H, Wu X, Liu X, Wang B, Wu Z, et al. Intra-abdominal hemorrhage following 739 consecutive pancreaticoduodenectomy: risk factors and treatments. *J Gastroenterol Hepatol*. (2019) 34(6):1100–7. doi: 10.1111/jgh.14560

50. Dhawan A, Lawlor MW, Mazariegos GV, McKiernan P, Squires JE, Strauss KA, et al. Disease burden of crigler-najjar syndrome: systematic review and future perspectives. *J Gastroenterol Hepatol*. (2020) 35(4):530–43. doi: 10.1111/jgh.14853

51. Akai M, Iwakawa K, Yasui Y, Yoshida Y, Kato T, Kitada K, et al. Hyperbilirubinemia as a predictor of severity of acute appendicitis. *J Int Med Res*. (2019) 47(8):3663–9. doi: 10.1177/0300060519856155

52. Lv T-R, Hu H-J, Regmi P, Liu F, Li F-Y. The effect of preoperative jaundice in the surgical management of gallbladder carcinoma: an updated meta-analysis. *ANZ J Surg*. (2021) 91(7–8):E455–464. doi: 10.1111/ans.17000

53. Ballowitz L, Hanefeld F, Schmid F. The influence of Various aminoglycoside preparations on bilirubin/albumin binding. *J Perinat Med*. (1976) 4(3):168–83. doi: 10.1515/jpme.1976.4.3.168

54. Kundur AR, Santhakumar AB, Bulmer AC, Singh I. Mildly elevated unconjugated bilirubin is associated with reduced platelet activation-related thrombogenesis and inflammation in gilbert's syndrome. *Platelets*. (2017) 28 (8):779–85. doi: 10.1080/09537104.2017.1280146

55. Kawakami H, Kondo S, Kuwatani M, Yamato H, Ehira N, Kudo T, et al. Preoperative biliary drainage for hilar cholangiocarcinoma: which stent should be selected? *J Hepato-Biliary-Pancreat Sci* (2011) 18(5):630–5. doi: 10.1007/s00534-011-0404-7

56. Wang Q, Gurusamy KS, Lin H, Xie X, Wang C. Preoperative biliary drainage for obstructive jaundice. *Cochrane Database Syst Rev*. (2008) (3):CD005444. doi: 10.1002/14651858.CD005444.pub2

57. Celotti A, Solaini L, Montori G, Coccolini F, Tognali D, Baiocchi G. Preoperative biliary drainage in hilar cholangiocarcinoma: systematic review and meta-analysis. *Eur J Surg Oncol J Eur Soc Surg Oncol Br Assoc Surg Oncol*. (2017) 43(9):1628–35. doi: 10.1016/j.ejso.2017.04.001

58. Atkinson M, Happey MG, Smiddy FG. Percutaneous transhepatic cholangiography. *Gut*. (1960) 1:357–65. doi: 10.1136/gut.1.4.357

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