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Preliminary results of *Moringa oleifera* Lam. grown in a semi-arid Mediterranean environment in a climate change scenario

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Introduction: Climate change, driven by greenhouse gas emissions, is altering global temperature and precipitation patterns, particularly affecting Mediterranean regions. Adaptation strategies, such as introducing low-input and resilient crops, are essential. *Moringa oleifera* Lam., a drought-tolerant tree native to north-west India, has emerged as a promising candidate due to its high nutritional value, rapid growth, and adaptability to arid environments.

Methods: This study evaluated the effects of planting methods and spacing on the growth and leaf nutrient composition of *M. oleifera* in Sicily (southern Italy), a semiarid Mediterranean environment. The experiment was conducted over two growing seasons (2021–2022). In 2021, four treatment plots were tested: two planting methods (seeding, S; transplanting, T) and two spacings (50 cm and 100 cm). In 2022, based on 2021 results, two plots were maintained to assess spacing effects (50 and 100 cm) under transplanting.

Results: Transplanted plants (T) showed higher values than seeded (S) in plant height and biomass production. The T50 treatment reached the highest leaf biomass (15 kg ha^{-1}) and nutrient accumulation. Total nitrogen content was 27 kg ha^{-1} in 2021 and 125 kg ha^{-1} in 2022 in T50. Similar trends were observed for phosphorus, calcium, potassium, and magnesium concentrations.

Discussion: Results demonstrate that *M. oleifera* maximizes growth and nutrient uptake when transplanted at 50 cm spacing. This cultivation approach supports its potential as a viable alternative crop in semiarid Mediterranean systems, promoting agricultural diversification and resilience for farmers.

KEYWORDS

leaf biomass, plant growth, macronutrients, mechanical transplanter, seeding, superfood, intercropping cultivation

1 Introduction

Climate change is causing global warming and an array of environmental challenges, including increased land desertification, pollution, and declines in biodiversity and agricultural productivity (Stocker et al., 2014; Thornton and Gerber, 2010; Bellarby et al., 2013; Gerber et al., 2013). According to the IPCC report (2022), all previous climate change assessments for the Mediterranean Basin and its subregions indicate ongoing warming of the atmosphere, as well as projected warming and changes in precipitation. The projected increase in climate risks, in combination with high regional vulnerability and exposure, make it an important "climate change hotspot." Within this context, small farms, in regions like the Sicilian hilly hinterland, are especially vulnerable. To overcome the vulnerability of these regions, adjustments in crops and cropping systems are needed, such as introducing new plants and cultivars, particularly superfoods.

Moringa oleifera Lam., due to its adaptability to different climatic conditions and diverse benefits, including nutritional, agricultural, medicinal, and environmental advantages (Ndubuaku et al., 2015; Daba, 2016), stands out as a promising candidate for supporting agriculture in semiarid Mediterranean environment.

Moringa oleifera Lam., well-known as 'drum-stick tree' or 'horseradish tree', is a fast-growing, evergreen or deciduous species which belongs to the Moringaceae family. The genus Moringa consists of 13 species, divided into three groups based on trunk shape. Bottle-shaped trees, tuberous shrubs and slender trees are distinguished. The latter includes *M. oleifera,* which is widely known and distributed due to its high ecological plasticity and adaptability to diverse climatic and soil conditions. This has led to its naturalization in tropical, subtropical, and arid climates. Other moringa species have a more limited distribution due to factors such as specific ecological requirements (Bania, 2023) or lesser-known and exploited uses. The studies by Korsor (2019) in a semi-arid environment and Dawit et al. (2016) in a tropical sub-humid environment compared the growth performance of *M. oleifera* with *M. ovalifolia* and *M. stenopetala*, respectively. In both studies, *M. oleifera* exhibited significantly greater height growth than the compared species, confirming its adaptive potential across diverse environmental conditions. Also, *M. oleifera* is common mainly in Middle East, African and Asian countries, but also in South and Middle American and Oceanian territories due to its ability to provide good yields (Prasad, 2011) along with its easy propagation (Bancessi et al., 2020), which is directly related to its suitable adaptation to climate change in regions where mild conditions have changed to arid ones, such as in the Mediterranean area.

Moringa oleifera performs better in climate with mean air temperature between 25 and 35°C but can tolerate temperatures up to 48°C in the shade and can survive a light frost. The drought-tolerant tree grows well in areas receiving annual rainfall amounts that range from 250 to 1,500 mm. *M. oleifera* thrives best at altitudes below 600 m; however, due to its high adaptability, it can also grow at elevations up to 1,200 m in tropical regions (Palada and Chang, 2003). Moringa is adapted to many types of soils, except those heavily clay or subject to waterlogging. Slightly calcareous clayey and sandy loam soils are considered the optimum media for this species due to their good drainage (Ramachandran et al., 1980; Abdul, 2007), with pH between 4.5 and 8. This species can tolerate water with an electrical conductivity (EC) of 3 dS m^{-1} during its germination phase, while at later stages its resistance to saline water increases (Oliveira et al., 2009). Moringa trees thrive in challenging soil and environmental conditions, requiring minimal maintenance while producing substantial biomass in various agroecological zones. Moringa is able to absorb water from deeper soil layers with its taproot system, ensuring survival during periods of drought when many commercial crops fail. Its high plasticity and stomatal conductance optimize physiological performance and growth under normal and water-stressed conditions by regulating the biosynthesis of secondary metabolites such as flavonoids and isoprenoids, which help mitigate photoinhibition and oxidative damage (Brunetti et al., 2020; Brunetti et al., 2018). Even under low rainfall, Moringa maintains a remarkable transpiration rate influenced by high temperatures, but under prolonged drought, it adapts by gradually reducing transpiration (Mabapa et al., 2018). It is also able to maintain a high relative leaf water content even under low soil moisture conditions, preserving cell physiological processes and growth (Rivas et al., 2013), allowing it to be grown in water-deficient regimes. In response to water deficit, it decreases stomatal conductance to minimize water loss (Mabapa et al., 2018; Munjonji et al., 2016). Studies conducted in arid and semi-arid regions of Tunisia indicate that water deficiency leads to reduced height growth and root elongation compared to control plants (Boumenjel et al., 2021), a phenomenon attributed to increased concentrations of growth-inhibiting regulators such as cytokinins and abscisic acid (Benmahioul et al., 2009). However, Moringa maintains high leaf yield even during short periods of drought. Nowadays, numerous studies and research are oriented toward the valorization and the use of plant species, both endemic and exotic, particularly rich in bioactive compounds, nutrients, and with high adaptation capacity to environmental stresses (Fascella et al., 2019; Fascella et al., 2023; Garofalo et al., 2024; Greco et al., 2024).

Moringa oleifera has been grown and consumed in its native areas by rural populations and the interest of scientists occurred only a few decades ago (the early 90s). Former research having Moringa as topic has been focused on its use in water treatments, because the seeds have proteins with cationic polyelectrolyte action, capable of neutralizing negatively charged colloids and can therefore be used as a non-toxic natural polypeptide for sedimenting mineral particles and organics (Foidl et al., 2001). Only later, its multiple uses and nutritional properties were investigated. Nowadays, Moringa cultivations are focused on leaf production. The leaves may be consumed as a salad, roasted, or stored as dried powder (Mammano et al., 2022a; Garofalo et al., 2024) for a long period without losing nutritious content (Garofalo et al., 2024). Moyo et al. (2011) determined the nutritional value of Moringa leaves using proximate and Van Soest methods. The dried leaves had crude protein levels as high as 30%, 19 different amino acids and many other mineral nutrients (Table 1). Moringa powder can be used as a substitute for iron tablets; its leaf powder has up to 28 mg kg^{-1} of iron (Gopalakrishnan, 2016), and up to 31 mg kg^{-1} of zinc (Barminas et al., 1998).

The effects of agronomic practices on leaf quantity production are crucial to standardizing production protocols, depending on the growing environment. However, the genetic resources of *M. oleifera* have not yet been adequately investigated and exploited for utilization, conservation, breeding, and improvement. Moreover, the success of crop improvement strategies depends largely on genetic characterization/assessment to identify potential parents for hybridization and crossing by heterosis (Ojuederie et al., 2014), which is lacking in *M. oleifera*. Leone et al. (2015) described various approaches to designing *M. oleifera* plantations for leaf production, depending on the intensity of the system adopted. In an intensive system, the plants are grown with a dense planting arrangement, with

TABLE 1 Nutritional values referred to 100 g of plant fresh and dried leaves and leaf powder of *Moringa oleifera* Lam. Fuglie (2001) and Moyo et al. (2011).

Nutrients	Fresh leaves	Dried leaves	Leaf powder
Calories (cal)	92	329	205
Crude protein (g)	6.7	29.4	27.1
Fat (g)	1.7	5.2	2.3
Carbohydrate (g)	12.5	41.2	38.2
Fiber (g)	0.9	12.5	19.2
Calcium (mg)	440	2,185	2003
Potassium (mg)	259	1,236	1,324
Iron (mg)	0.85	25.6	28.2
Magnesium (mg)	42	448	368
Phosphorus (mg)	70	252	204
Copper (mg)	0.07	0.49	0.57
Sulfur (mg)	–	–	870
Vitamin A (mg)	1.28	3.63	16.3
Vitamin B1 (mg)	0.06	2.02	2.64
Vitamin B2 (mg)	0.05	21.3	20.5
Vitamin B3 (mg)	0.8	7.6	8.2
Vitamin C (mg)	220	15.8	17.3
Vitamin E (mg)	448	10.8	113

spacing ranging from 10 cm × 10 cm to 20 cm × 20 cm, and a 35 to 45 days harvest interval. In this case, irrigation and fertilization are essential to sustain productivity. Semi-intensive production, on the other hand, involves wider spacing, approximately 50 cm × 100 cm, with harvesting occurring every 50–60 days, while irrigation and fertilization are still recommended. Finally, cultivation can be integrated into an agroforestry system, with row spacing of 2–4 meters, a harvesting interval of around 60 days, and reduced requirements for fertilization and irrigation, which are not strictly necessary.

According to Zheng et al. (2016) the highest planting density of 20 cm × 20 cm (approximately 250,000 plants ha^{-1}) in combination with the 30 cm cutting height produced the highest fresh matter (approximately 70 t ha^{-1}) and dry matter (approximately 12 t ha^{-1}) yield in the second evaluation, in southwest China. In Nicaragua, Mendieta-Araica et al. (2013) found that the planting densities of 167,000 plants ha^{-1} was compared to density 100,000 plants ha^{-1}, due to the possibility of achieving very high dry matter yield (19 t ha^{-1}) with a high proportion of fine fraction yield. Němec et al. (2020) reported that a plantation counting 705 of *M. oleifera* and 705 of *M. stenopetala* trees with a 100 × 150 cm current spacing shows a higher yield of leaf biomass than the expected yield if the spacing were changed to 300 × 300 cm. Although high densities are positively correlated with high dry matter yields, the spatial arrangement in the field, the high amount of labor needed and difficulties during harvesting make high densities impractical for small and medium-scale farmers (Mendieta-Araica et al., 2013).

Sicily is the largest island of Italy, located in the middle of the Mediterranean Sea, in which climate is characterized by hot summers, mild winters, and very changing middle seasons. Rainfall is generally scarce in late spring and summer, proving to be insufficient to ensure water supply in some areas. Although agricultural activity in Sicily has historically been characterized by the presence of olives, citrus, vines, forages, and other temperate climate crops, the region has been experiencing the effect of climate change in recent decades. Changes in temperature and rainfall patterns due to drought, excessive heat with temperatures that can rise beyond 30°C on more consecutive days, and unpredictable rainfall are the biggest concerns for Sicilian farmers. Climatic changes may affect the adaptability of temperate fruit trees that account for about 48% of global fruit production (Salama et al., 2021). In this context, to tackle these challenges, adaptation strategies and conservative agriculture practices are needed to ensure stable production over time and counteract production instability. One possible solution might be the introduction of species tolerating warmer climates, which in the context of global warming and changing production systems might require less input.

Moringa is a plant capable of adapting to the effects of climate change in Sicily, integrating into Mediterranean farming systems as an intercrop and forage crop, providing additional and diversified income for farmers.

In Sicily, *M. oleifera* could be part of the typical cultivation systems of the Mediterranean areas, both as a catch and as a forage crop. If planted in June and harvested in October–November, it could be considered a summer-autumn intercrop with cereals and fodder such as *Hedysarum coronarium* L.

Therefore, this study aims to evaluate the effect of planting mode and distances on leaf production and nutrient content of *M. oleifera* grown in this semiarid environment. This study represents one of the first attempts to evaluate the cultivation of Moringa in a semi-arid Mediterranean environment, aiming to fill the knowledge gap on the species' cultivation practices in this context and to exploit its potential as a low-input species.

2 Materials and methods

2.1 Study area and experimental design

The experiment was conducted over two consecutive years, in 2021 and 2022, at the "Basile Giuseppe" farm located in Ventimiglia di Sicilia (37.971203 N, 13.527763 E; Palermo, Sicily, Italy), in an open field previously used for sheep grazing. It is a model of Sicilian environmentally friendly multifunctional farm for soil protection (Mammano et al., 2022b).

The field is located at an elevation of 550 m above sea level and its soil type is Vertic Haploxerept (Soil Survey Staff, 2022) on clay parent material. The main physical and chemical properties of the soil (Table 1) were determined at the beginning of the study, with sampling at two depths (0–20 cm and 20–40 cm).

For the climatic analysis of the site, a thermo-pluviometric station was installed in the field. The ombrothermal diagram was made, using the data registered in 2021 and 2022, which are shown in Figure 1. From the same, it is possible to identify the months with rainfall deficit that constitute the dry period.

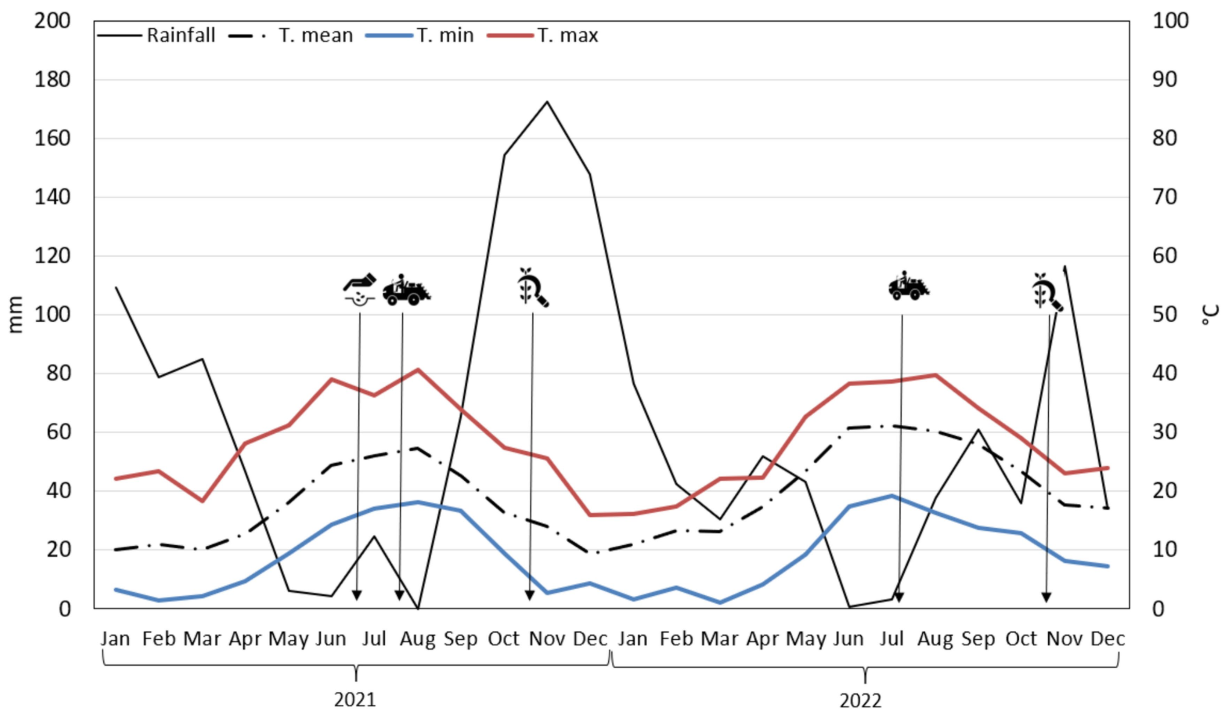

FIGURE 1
Ombrothermal diagram related to 2021 and 2022 years in field stations.

In particular, the diagram shows that the area on average is characterized by four dry months in 2021 and six dry months in 2022, thus attributable from the Mediterranean to Semiarid climate type, according to Köppen climate classification.

Referring to Peguy's climogram (Figure 2), which reports the trend of temperature and precipitation through a polygonal shape, it is found that this is characterized by a shape that develops mainly in a horizontal direction, which denotes a high seasonal temperature range, with precipitation accumulations occurring precisely in the autumn-winter period and completely scarce if not entirely absent in the spring–summer period. In general, the climate is arid from May to September and temperate during the remaining part of the year. From the climogram, it is observed the period January–April and October–December are closed in the Temperate climate. While May–September months are included in the Arid-Warm climate.

The experimental field covered an area of 980 m^2, of which 900 m^2 were considered for *M. oleifera* tests (Figure 3). The remaining 80 m^2 comprised the borders to facilitate management and cultivation practices. During the first cultivation cycle, in 2021, nine rows with four plots were set to test the effect of experimental factors. Specifically, two planting modes (S, seeding; T, transplanting) and two spacing levels on the row (50 and 100 cm) were evaluated, with uniform spacing between the rows of 200 cm. The treatments were selected based on literature studies (e.g., Gandji et al., 2018; Mathenge, 2015; Palada, 1996) regarding the most commonly used propagation methods for *M. oleifera*, which required validation under Mediterranean environmental conditions. Additionally, planting distances of 50 and 100 cm within the row were selected to assess growth capacity and leaf production in low-input and non-intensive systems, being more convenient for small to medium-sized Sicilian farms, as recommended by Mendieta-Araica et al. (2013). In 2022, based on results obtained in 2021, nine rows with two plots were set to assess the effect of planting distance (50 and 100 cm) on transplanted plants. In both years, the spacing distances evaluated were 50 cm x 200 cm and 100 cm x 200 cm, and the plant densities considered were 10,000 and 5,000 p ha^{-1}. These plots are verified for the Moringa mechanical harvesting (Comparetti et al., 2022).

2.2 Plant material and growth conditions

To obtain seedlings for transplanting, Moringa seeds were provided by "Giuseppe Morreale" agricultural farm located in Grotte (Agrigento, Sicily, Italy) and partner of the funding project "PREVANIA," and used by the Research Center for Plant Protection and Certification of Bagheria (Palermo, Sicily, Italy) for the multiplication phase to achieve a high germination rate. Seeds were obtained from parent plants grown in the "Giuseppe Morreale" nursery, harvested in November 2020 and kept at dark and dry, until seedling preparation operations for transplanting, in late May 2021 and direct seeding in mid-June 2021 (Figure 1). Moringa seeds were soaked in sterilized water for 48 h, placed subsequently in Petri dishes containing water-soaked filter paper and grown in a climatic cell at 23°C with 16 h photoperiod light. The germination rate was 95%.

Germinated seeds, having emergence of the plumule and rootlet, were successively placed in 10-cm-diameter pots

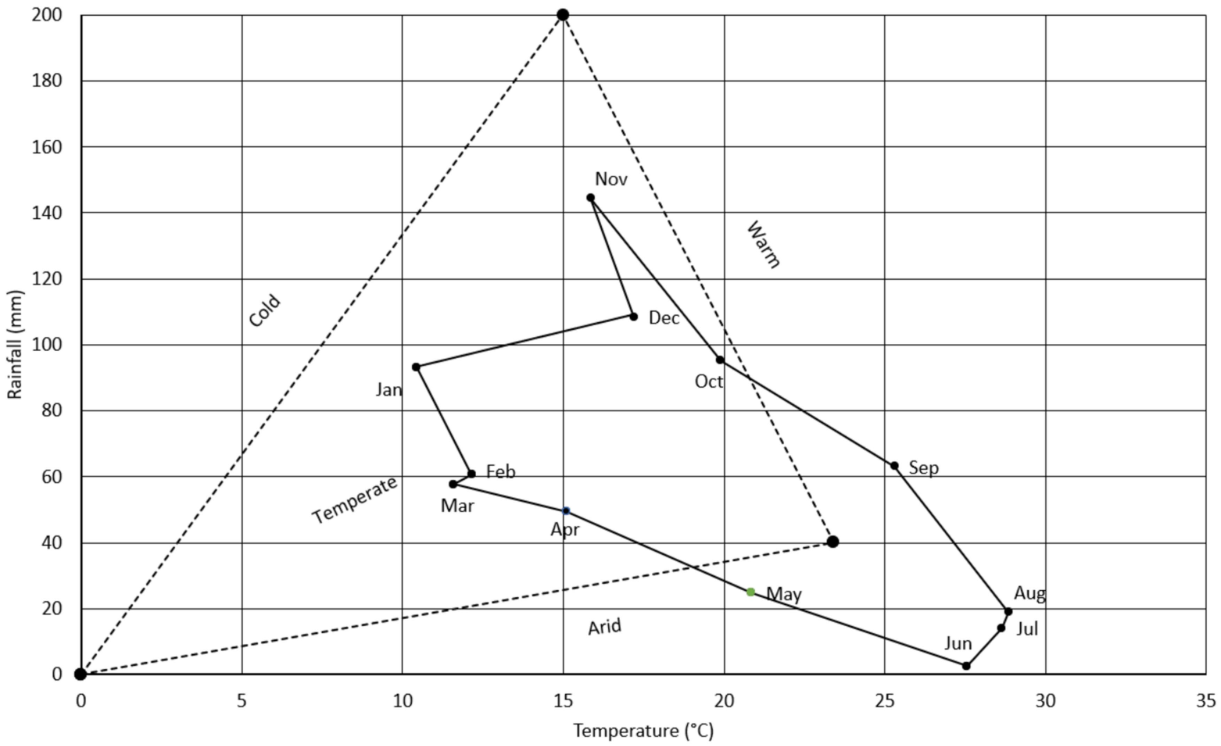

FIGURE 2
Péguy climographs obtained by averaging the monthly data from the thermometric and rainfall in field stations.

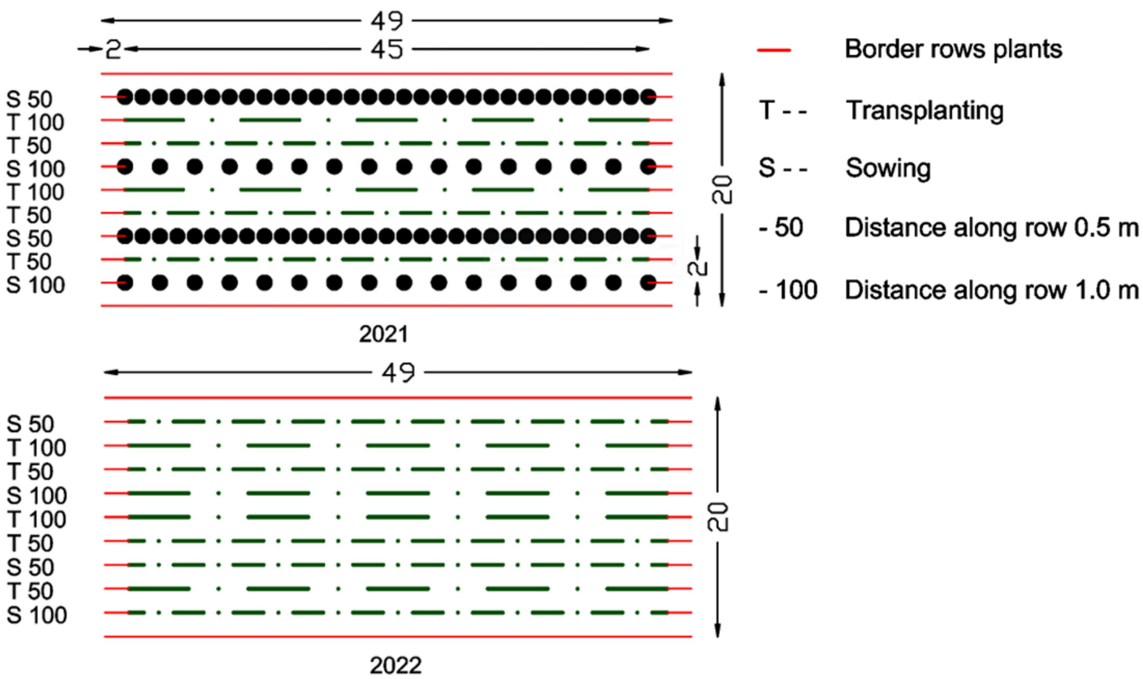

FIGURE 3
Experimental design of *Moringa oleifera* field test.

containing a growing substrate composed by a mixture of brown and blond peat to which 10% perlite was added. The pots containing the germinated seeds were placed in 50% shade netting.

Irrigation was carried out, three times a week, in order to keep the humidity of the growing substrate constant. The Moringa seedlings after 30 days reached a height of about 20 cm and a well-developed root system ready for transplanting to open fields (Figure 1).

2.3 Soil–plant system management

To prepare the field for planting and transplanting operations, the field was subjected to surface tillage to remove weeds and prepare seedbed for planting. A perforated pipe with a 16 mm diameter was installed along each row to facilitate supplementary irrigation. Fifty days after planting, inter-row surface tillage was performed to control weed growth and soil connectivity (Conte et al., 2017). No organic or mineral fertilization was carried out during the experiment.

On June 21st, 2021, direct hand seeding was performed sowing the seeds at a soil depth of 2 cm, using 2 seeds per hole (Figure 1). The germination rate was 91% and the first shoot was visible 20 days after the seeding. On July 27th, 2021 transplanting with a mechanical transplanter (Figure 4), was carried out, using seedlings with appropriate height, stem size and root dimensions. Following direct seeding and transplanting operations, localized irrigation was carried out to promote seedling germination and rooting, respectively. Harvest occurred on October 23rd, 2021 after a cultivation cycle of 17 weeks in order to compare the biometric growth, leaf yield, and macro- and micronutrient content of the four treatments under study, and to identify which of them provides the best performance in a semi-arid Mediterranean environment.

In 2022, a new cultivation cycle was established with the transplanting performed on July 4th, and plants harvest on October 24th, after 16 weeks of cultivation.

A Cecchi & Magli model Unifox transplanter was used in this work. It is a universal transplanter suitable for bare-root seedlings, conical, pyramidal and cubic-shaped clods. Because of its flexibility, it was chosen for moringa transplanting (Figure 4). This represents the first application of a Moringa transplanter, aimed at mechanizing the cultivation operations of Moringa across large agricultural areas.

A reinforced anti-wear plow (Figure 4; A) with depth adjustment (B) allows for the opening of a furrow where the moringa seedlings are deposited with a ball of earth using a clamp distributor (C). It is driven by the compacting support wheels (D) through a chain transmission system with replaceable sprockets (E) that enable different transmission ratios and variations of the spacing along the row. An operator seated on the seat (F) takes the seedlings with the ball of earth from the plant holder drawer (G) and places them in the still-open clamps (C), which, while advancing, hold the seedling to subsequently release it into the previously opened furrow. The hilling tools (H) allow for the anchoring of the seedlings to the ground and the closure of the previously opened furrow. The compacting wheels (D) can perform optimal hilling. The pressure exerted by the compacting wheels can be adjusted through a spring (I) and adjustable links (L) that change the position of the wheels (M) relative to the frame. The machine is connected to the tractor via a three-point hitch (N). Before transplanting, the machine was specifically adjusted in the field to verify the correct spacing for depositing the seedlings along the row.

2.4 Plant measurement and leaf harvest

In both years of the trial, on the day of harvest, plant height and stem diameter at the collar level were recorded to assess the vegetative growth. Separation of the main stem and secondary branches from the leaf and tertiary herbaceous branches biomass was carried out manually. The fresh weight of the leaves from each plant in each

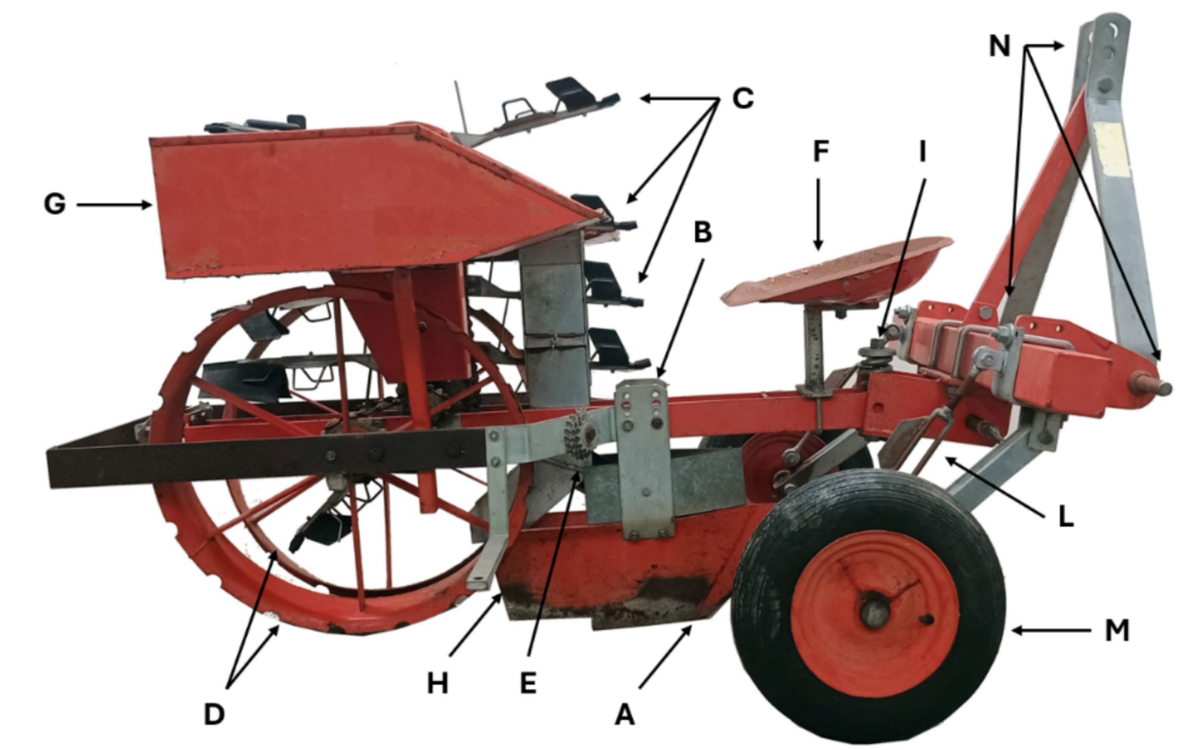

FIGURE 4
Mechanical transplanter used in the field tests.

treatment was recorded immediately after harvesting using an electronic balance. Subsequently, all fresh biomass for each treatment was placed on raised pallets inside a shaded and ventilated greenhouse, in a controlled environment, until completely air dried. The dried leaves were separated from the tertiary branches and finely ground into a powder, subjected to ozone treatment to reduce the surface microbial load, and used for nutrient content analysis.

2.5 Nutrient content of *Moringa oleifera* leaves

Nitrogen (N), phosphorus (P), calcium (Ca), magnesium (Mg), potassium (K), sodium (Na) and iron (Fe) concentrations of *M. oleifera* leaves were determined at the end of both harvesting periods on ground dried leaves samples. To determine the concentration of mineral nutrients, leaves powder was firstly pre-digested with concentrated nitric acid (HNO_3) and hydrogen peroxide (H_2O_2) in porcelain capsules placed in hot (250°C) sand bath. After evaporation of nitric acid and hydrogen peroxide, samples were mineralized in a muffle at 550°C. The obtained ashes were suspended with nitric acid at 2% (v/v) and analyzed by Microwave Plasma Atomic Emission Spectroscopy (Agilent 4,210 MP-AES, Milan, Italy).

Moringa oleifera leaves were pulverized in a porcelain mortar, and the resulting ground materials (0.25 g) underwent mineralization in porcelain crucibles within a muffle furnace at 550°C for 8 h.

The resulting ashes were suspended by using 1 M HCl on a hotplate at 100°C for 10 min. The digested samples were recovered in 15 mL tubes and adjusted to 10 mL of volume with MilliQ grade water. P concentration was then determined using the colorimetric method (Murphy and Riley, 1962) using a spectrophotometer (UVmini-1240, Shimadzu Italia srl, Milan, Italy) with ammonium molybdate solution (Murphy and Riley, 1962).

Total N was determined by the Kjeldahl method (Bremner, 1996). Briefly, 2 grams of powdered leaves were weighed in pyrex digestion tubes and digested at 360°C for 3 h with concentrated sulfuric acid (98% H_2SO_4) and copper oxide as catalyzer. After the digestion, ammonium was recovered by Kjeldahl distillation using 3% (v/v) of boric acid (H3BO3). Total N was assessed by automatic titration of H_3BO_3 with diluted (0.5 M) H_2SO_4.

2.6 Statistical analysis

Reported values are the arithmetic mean ± standard deviations of three replicates. Before performing parametric statistical analyses, normal distribution and variance homogeneity of the data were checked by Shapiro-Wilks test and Levene's tests, respectively (p-value >0.05). Data of the first year of the experiment (2021) were subjected to Two-way ANOVA, considering the planting mode (seeding (S) or transplanting (t)) and the distance (50 or 100 cm). Data of the second year of the experiment were subjected to ONE-WAY ANOVA with planting mode (S, seeding; T, transplanting) as factor. Mean significant differences were assessed by Tukey post-hoc test or -student test at $p < 0.05$. All the statistical analyses were performed using SPSS 13.0 for Windows (SPSS Inc. 1996).

3 Results and discussion

3.1 Plant growth parameters

In 2021, harvesting was done by cutting the central stem 40 cm above ground level to test whether the plants had the ability to regrow in a Mediterranean environment. Probably, due to low winter temperatures, the plants showed no ability to regrow. In addition, seeding produced stunted and small plants, so it was decided not to repeat the treatment and focus on the growth potential of the transplant. Plants obtained through direct seeding did not exhibit adequate development or adaptability, making this propagation method unsuitable for establishing a commercial plantation. The planting distances, selected to reduce input use and promote environmental sustainability, further highlighted the limitations of seed-derived plants, which proved to be less competitive and more vulnerable. This would also lead to increased costs for farmers interested in investing in the species, making direct seeding a less viable option from an economic standpoint. Therefore, in the second year of the study, the focus shifted to identifying the most effective treatments in terms of plant development and leaf production. In 2022, the cultivation cycle was established with newly transplanted plants, which were ultimately harvested by cutting the main stem 20 cm above ground level.

The results reported in Table 2 show significant differences in plant size of transplant compared with seeding and a clear growth trend for transplanted plants.

In 2021, a highly significant difference was found when comparing treatments with the same spacing (S50, T50; S100, T100), although within the planting treatment (S50, S100; T50, T100) no significant changes in height and diameter were observed, at 17 weeks after planting. The transplanted plants showed higher values in terms of height and diameter at both planting distances than their seeding counterparts, with the T100 showing the greatest growth in height and diameter (350 cm, 4.5 cm; Table 2).

This different behavior, between seeding and transplanting, can be related to the different growth capacities of plants originating from direct seeding and transplanted plants. It is known that the applicability and success of direct seeding depend on multiple factors such as seed quality, knowledge of appropriate sowing times and rates, seedbed preparation, field germination, emergence, and resource competition from emerging weeds (Douglas et al., 2007). In addition, the main limiting factors of direct seeding are uneven stands, disease, weed competition, insufficient nutrients, and temporary or ongoing environmental challenges (such as drought, drying events, flooding and extreme heat). Conversely, many authors agree that transplanting is a reliable method to improve growth and achieve earliness and higher yield, as reported for several crops, including tomato, rice, bell pepper and watermelon (Ketema et al., 2013; Hossain et al., 2002; Leskovar and Cantliffe, 1994). According to Palma and Laurance (2015), using direct seeding and seedling plantings in restoration projects revealed that the successful survival percentage of transplants in different plant species was three times higher than direct seeding. Traditionally, in Sudan seeds are preferred; while vegetative propagation is common in India, Indonesia and in some areas of West Africa (Leone et al., 2015; Palada, 1996).

TABLE 2 Physical and chemical properties of the soil, at two depths (0–20 cm and 20–40 cm), used in the experiment.

Property	Measurement units	0–20 cm	20–40 cm
Sand	%	36	38
Silt	%	27	27
Clay	%	37	35
pH		7.78	7.77
Electrical Conductivity	$\mu S\ cm^{-1}$	861.3	821.9
Total Organic Carbon	%	5.0	5.0
Total Nitrogen	$g\ kg^{-1}$	3.3	3.0
Available P	$mg\ kg^{-1}$	43	39
Cation Exchange Capacity	$cmol_+\ kg^{-1}$	28	28

In 2022, T100 is the more effective treatment for plant growth than T50, reaching significant values of 334 cm height and 4.1 cm diameter, at 16 weeks after planting. Compared with the previous year, both treatments showed a slight decrease in growth trend. This could be attributed to harvesting a week earlier than the previous year, or alternatively, a possible resource depletion in the soil. Thus, according to Mabapa et al. (2017), it is advisable to apply fertilizers for improving its growth and yield in areas with low rainfall and extreme temperatures. It was found that poultry manure produced better stem girth and vegetative growth than NPK fertilizer (Dania et al., 2014) making it possible to reduce the excessive use of chemical fertilizers, which are known to have negative effects on the environment.

However, the results indicate that to maximize plant growth, it is useful to adopt wider planting distances (100 cm x 200 cm) than narrower planting distances (50 cm x 200 cm) and to use transplanted plants. The findings of Yilma et al. (2022), confirm that, in *Moringa stenopetala* grown in Ethiopia, medium (100 cm x 100 cm) and wider spacing (200 cm x 200 cm) gave significantly higher average height growth than narrow spacing of 50 × 50 cm at the 6, 9, 12 and 15th months after planting. Similar patterns were observed in *M. oleifera*, where plants at wider spacing performed better in terms of growth in height and stem diameter, in southern Africa by Olawepo et al. (2020), and in West Africa by Goss (2012). Therefore, a study conducted by Mashamaite et al. (2021), in Limpopo Province (South Africa), which has a similar climate similar to the Mediterranean, shows that high density is ideal for optimum leaf production and the planting density considered in our work, can be regarded as medium, such that it is conducive to the livelihood and well-being of farmers and does not excessively deplete soil resources.

3.2 Leaf biomass production

In 2021, after separation of main stem and secondary branches, fresh biomass production included leaves and herbaceous tertiary branches. The average fresh biomass production reached 7 t ha^{-1}, 5.5 t ha^{-1}, 34 t ha^{-1} and 24 t ha^{-1} for S50, S100, T50 and T100 treatments, respectively. In 2022, the average fresh biomass production values were 37 t ha^{-1} and 25 t ha^{-1} for T50 and T100. As a result of the drying process, a loss of water content of 18% in the first year, and of 19% in the second year was assessed. The dry leaf component of the obtained biomass accounted for 40 and 38%, in 2021 and 2022, respectively. Thus, the production in dry leaf biomass (Figure 5) was 0.5 t ha^{-1}, 0.4 t ha^{-1}, 2.5 t ha^{-1} and 1.7 t ha^{-1} for S50, S100, T50 and T100 treatments in 2021, and 2.7 t ha^{-1} and 1.8 t ha^{-1} for T50 and T100 in 2022, respectively.

Based on these results, a narrower planting density produced a higher leaf biomass yield than a wider spacing. This is valid for both sowing and transplanting, even if the yields of the sowing treatment are not considered satisfactory. In transplant treatments, however, an inverse correlation is observed between height, diameter of the plants and production of dry biomass.

Increased height and diameter of individual trees as a result of increased spacing (T100) did not contribute to the production of significantly higher dry leaf biomass per plant. This could be due to less competition between plants that led to a different use of resources, which may have been directed to root system development, diameter growth or longer internodes (which was not recorded), rather than leaf production. However, the increased growth of individual trees due to increased spacing was not sufficient to offset the effect of reduced density per hectare on dry leaf biomass. For narrower planting distances (T50), the lower yield potential of a single plant is balanced by the larger plant population. Suitable plant density is essential for establishing adequate leaf area for light interception, photosynthesis, and ultimately, maximum crop development and yield to achieve a high biomass production. Other studies reported a positive linear relationship between planting density and biomass yield in *M. oleifera*. Goss (2012) indicated that the highest population density (197,528 plants ha^{-1}) spaced at 25 cm x 25 cm and the second highest population density (98,764 plants ha^{-1}) spaced at 35 cm x 35 cm, produced the highest leaf fresh weight figures of 404 g $plot^{-1}$ and 205 g $plot^{-1}$, respectively.

Patricio et al. (2015) carried out a study comparing different planting densities and harvest intervals. The results show that overall, fresh leaf biomass increased as the plant population increased from 10,000 to 40,000 plants ha^{-1}. The maximum density of 40,000 plants ha^{-1} resulted in a high leaf biomass yield, while a longer harvest frequency (8-week interval) produced a higher yield than shorter intervals. Findings from previous studies conducted concurrently with this one, such as Mabapa et al. (2017) and Amaglo et al. (2006), showed that the plants' greatest output of leaf biomass per hectare was obtained at the closest experimental spacing.

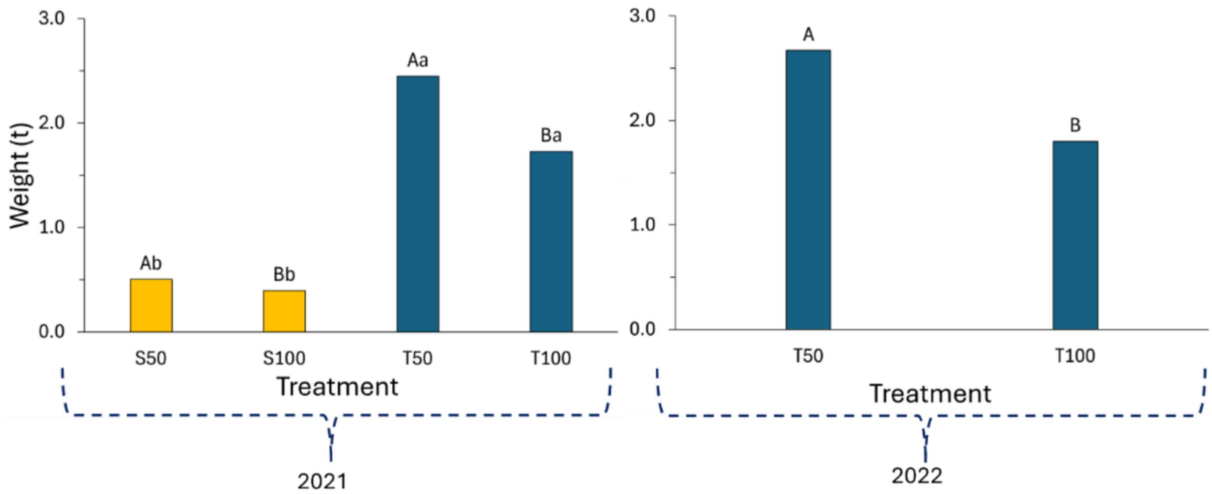

FIGURE 5
Dry weight (t) of *M. oleifera* dried leaves at the end of each experimental year (2021–2022), planted using seeding (S) and transplanted (T) at two distances (50 and 100 cm). Different capital letters indicate significant differences (p < 0.05) between the two distances (50 and 100 cm) within the same planting mode (S or T). Different lowercase letters indicate significant differences (p < 0.05) between the two planting modes (S or T) at the same distance (50 and 100 cm). Reported results are expressed as mean ± standard deviation of three field replicates.

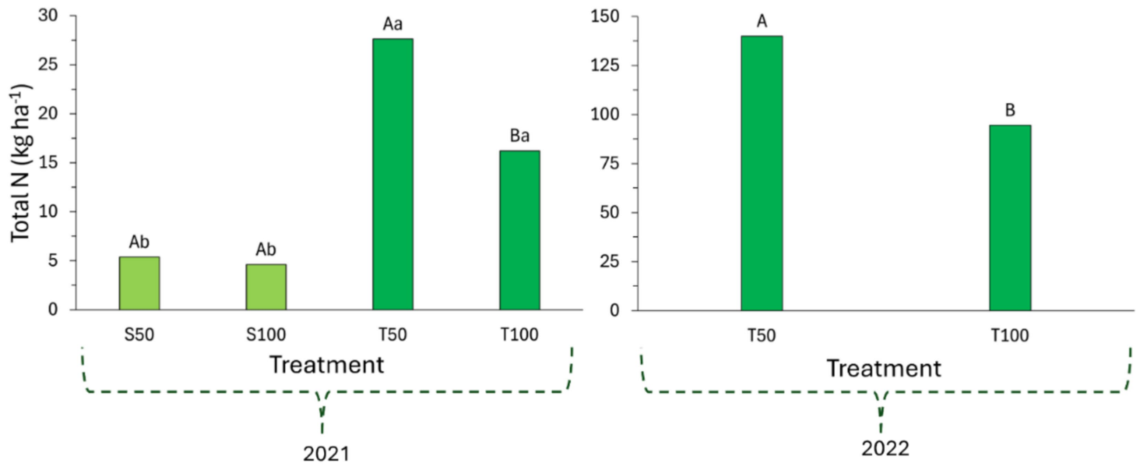

FIGURE 6
Total nitrogen in *M. oleifera* dried leaves at the end of each experimental year (2021–2022), planted using seeding (S) and transplanted (T) at two distances (50 and 100 cm). Different capital letters indicate significant differences (p < 0.05) between the two distances (50 and 100 cm) within the same planting mode (S or T). Different lowercase letters indicate significant differences (p < 0.05) between the two planting modes (S or T) at the same distance (50 and 100 cm). Reported results are expressed as mean ± standard deviation of three field replicates.

3.3 Total nitrogen and phosphorus contents in *Moringa oleifera* leaves

In 2021, treatments S50 and S100 showed relatively low nitrogen (N) content (Figure 6), around 10 kg ha^{-1}, with no significant differences between them. On the contrary, the T50 and T100 treatments showed significantly higher N content. T50 reached a value close to 27 kg ha^{-1}, while T100 stood at around 17.5 kg ha^{-1}. A significant rise in the N content is also noticed for both T treatments for the year 2022 in relation to the values of the previous year. T50 recorded an N content above 125 kg ha^{-1}, while T100 reached around 100 kg ha^{-1} (Figure 6). The comparison between 2021 and 2022 reveals a significant increase of N levels in both T treatments. These results could be attributable to the previous use of the field as pasture and the presence of crop residues from the 2021 cycle, which may have contributed to increased mineralization and nitrogen availability in the second year.

Other studies have examined protein content of Moringa in semi-arid growing environment. Abbas et al. (2018) examined the protein content in dried *M. oleifera* leaves grown in Sudan. According to their results, the protein values stood at 8%, a level comparable to that obtained in this experiment, especially in 2021, where levels ranged from 6% in T100 to 7% in S100. Yaméogo et al. (2011) evaluated the protein content in dried *M. oleifera* leaves cultivated in three different areas, grown in the semi-arid environment of Burkina Faso. These latter results showed protein values of 27.6%, quite comparable with

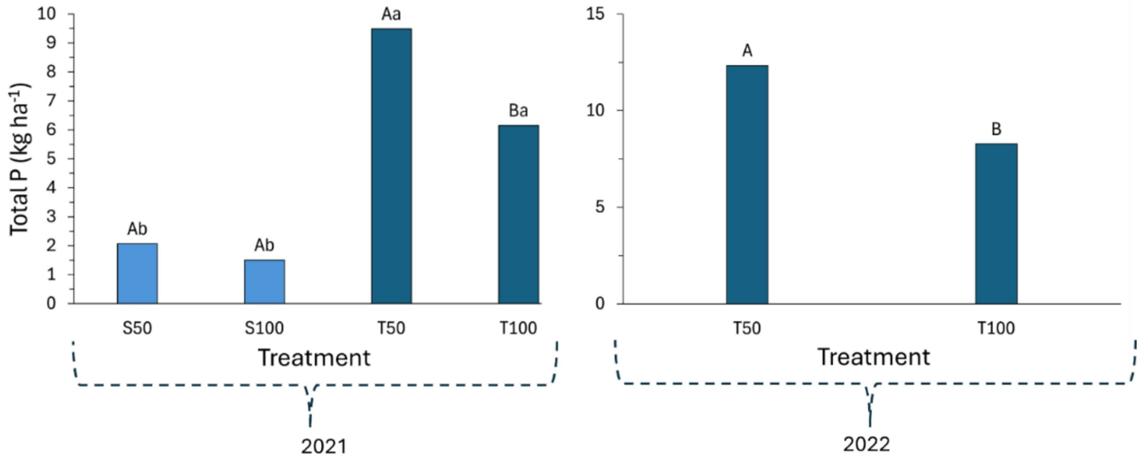

FIGURE 7
Total phosphorus in *M. oleifera* dried leaves at the end of each experimental year (2021–2022), planted using seeding (S) and transplanted (T) at two distances (50 and 100 cm). Different capital letters indicate significant differences ($p < 0.05$) between the two distances (50 and 100 cm) within the same planting mode (S or T). Different lowercase letters indicate significant differences ($p < 0.05$) between the two planting modes (S or T) at the same distance (50 and 100 cm). Reported results are expressed as mean ± standard deviation of three field replicates.

the results of our study, especially in both 2022 transplant treatments, where values of about 32% were reached (Figure 6).

Analysis of total phosphorus (P) content in *M. oleifera* leaves (Figure 7) shows different trends across treatments and years. Within 2021, the S50 and S100 treatments showed similar total P contents of about 2 kg ha^{-1} and 3 kg ha^{-1}, respectively, with no statistically significant differences. In contrast, the T50 and T100 treatments revealed significantly higher P contents, with T50 reaching about 9 kg ha^{-1}, while T100 recorded a value of about 7 kg ha^{-1} indicating that T50 is the most effective treatment in P accumulation in 2021. In 2022, T50 reached significantly higher P content (12 kg ha^{-1}) than T100 (8 kg ha^{-1}; Figure 7). The maintenance of high phosphorus contents, even in the absence of fertilization may be due to that these planting densities did not induce any competition among the plants, such that phosphorus uptake was not prevented, which is known to be a weakly mobile element.

Overall, the results of the last two analyses suggest that transplanting, especially at a spacing of 50 cm, is the most effective strategy for maximizing total N and P accumulation in *M. oleifera* leaves.

The results of this experiment are in line with other similar studies that have examined the P content in dried Moringa leaves. According to Sultana (2020), the highest P concentration in Moringa leaves reached 0.3%, similar to the concentration obtained in the transplant treatments of both years of our experiment, with a peak of 0.37% in T50 of 2021 and a maximum of 0.43% in T50 of 2022. Yaméogo et al. (2011) found P concentrations ranging from 0.43 to 0.25%. These latter results are comparable to those achieved in the present study, specifically the T50 treatment in 2022 reached P concentrations of 0.43%, followed by T100 in 2022 which reached 0.4%, and T50 in 2021 with 0.37% (Figure 7).

3.4 Total mineral nutrient contents in *Moringa oleifera* leaves

The results of the analysis, shown in Table 3, reveal a clear comparison between the different treatments (S50, S100, T50, and T100) in the 2 years considered, showing significant changes in mineral nutrient levels. In 2021, mineral content varies widely by planting mode treatment. The transplant treatments showed high content in both planting distances (T50 and T100), this supposes that planting mode affects mineral nutrient accumulation in Moringa plants. Non-significant differences were recorded in the seeding treatments. T50 and T100 produced the highest mineral nutrient contents for K, Na and Fe, while T50 recorded the highest values for Ca content with 149.0 kg ha^{-1} and Mg with 23.9 kg ha^{-1} (Table 3).

In 2022, there is an overall decrease in mineral nutrient accumulation compared to 2021, with T50 maintaining significantly higher values for Ca (67.4 kg ha^{-1}), K (69.6 kg ha^{-1}) and Mg (12.2 kg ha^{-1}) content compared to those from T100 (44.7 kg ha^{-1}, 45.2 kg ha^{-1} and 7.1 kg ha^{-1}, respectively). On the other hand, Na

TABLE 3 Height and diameter of *M. oleifera* plants at the end of each experimental year (2021–2022), planted using seeding (S) and transplanted (T) at two distances (50 and 100 cm).

Treatment	Height	Diameter
2021		
cm		
S50	89 Ab	0.9 Bb
S100	72 Ab	1.7 Ab
T50	280 Aa	3.7 Ba
T100	350 Aa	4.5 Aa
2022		
cm		
T50	273 A	3.6 A
T100	334 b	4.1 B

Different capital letters indicate significant differences ($p < 0.05$) between the two distances within the same planting mode. Different lowercase letters indicate significant differences ($p < 0.05$) between the two planting modes at the same distance. Reported results are expressed as mean ± standard deviation of three field replicates.

values for both treatments decrease significantly compared to 2021, but no significant differences emerge between T50 and T100 (Table 3). Finally, there is an overall Fe decrease from 2021, with T50 remaining higher (0.4 kg ha^{-1}) than T100 (0.2 kg ha^{-1}). This general decrease in mineral nutrient accumulation, between the two experimental years, could be due to the non-application of any fertilization or input of organic matter to the soil, during the year, which led to a slight depletion of the substrate, especially in micro-nutrients.

The results of our analysis showed that Ca, K and Mg are the elements found in the highest concentration in *M. oleifera* leaves grown in the semiarid Mediterranean environment. The present results agree with Leone et al. (2015) who reported that Ca, K and Mg are the predominant minerals in the *M. oleifera* tissues. In particular, the highest content of K is found in the vegetative parts and immature pods, while leaves, and seeds are rich in Ca and Mg, respectively. Other studies have analyzed the mineral concentration of dried *M. oleifera* leaves. The results of Teixeira et al. (2014) partially agree with those of this study, regarding the Ca concentration that was 3%, while in our experiment Moringa leaves reached concentrations of 6% in the T50 and S100 treatments of 2021. Sultana (2020) also reported similar Ca values of 2.64%, which is in line with the 2022 transplant results of 2.25% for T50 and 2.48% for T100 (Table 3).

The K content in our study was similar to the results of Sultana (2020) (2.02%), especially for S100 in 2021 with 2.75% and in T50 in 2022 with 2.6%. The Mg concentration of Moringa reached a maximum of 1% in the T50 treatment in 2021, which is intermediate between the results obtained by Teixeira et al. (2014) and Moyo et al. (2011) of 2 and 0.5%, respectively (Table 4).

4 Conclusion

Moringa oleifera is increasingly recognized for its resilience and adaptability, making it a promising alternative crop for Sicilian farmers facing the challenges of climate change. As traditional crops struggle with rising temperatures, erratic rainfall, and soil degradation, Moringa thrives in diverse conditions, requiring minimal water and being tolerant to drought. This characteristic positions it is an ideal candidate for cultivation in regions as Sicily where water scarcity is becoming a pressing issue.

Moreover, the nutritional value of Moringa is exceptional. Its leaves are rich in vitamins, minerals, and antioxidants, making it a sought-after superfood in both local and international markets. The global demand for health-conscious products is on the rise, and Moringa could offer Sicilian farmers an opportunity to tap into this lucrative market. By diversifying their crop portfolio to include Moringa, farmers can not only enhance their income but also reduce their reliance on traditional crops that may be less viable under changing climatic conditions.

This is the first study that evaluated *M. oleifera* Lam. cultivation in a semi-arid Mediterranean environment. This trial assessed vegetative growth potential, leaf biomass production, and nutrient accumulation in *M. oleifera* leaves, grown in Sicily, in two consecutive years.

This study was established with a focus on environmental sustainability and input reduction in agricultural systems. The selection of wider planting distances, to reduce irrigation inputs, and no fertilization during the experimental trial are part of this approach.

Results showed that transplanted plants achieved higher values of height than seeded plants, as well as a leaf biomass production of 2.8 t ha^{-1} in the T50 treatment of 2022. Total N content was higher in the T50 treatment, reaching 27 kg ha^{-1} in 2021 and a peak of 125 kg ha^{-1} in 2022. Similarly, the T50 treatment recorded the highest levels of total P, Ca, K, and Mg. This study reveals that the combination of factors that maximize leaf biomass production and the greatest accumulation of macronutrients in leaves, in both years of the experiment, was transplanting at 50 cm (T50), with values at least twice as high as T100 and S50.

Our results could help to define low-input and eco-friendly cultivation protocols of Moringa with practical implications, from an environmental and an economic point of view as involve water-fertilizers saving and minimum tillage practices, for farmers working in the semiarid areas of south Mediterranean basin. Future studies of this species should be addresses on the evaluation of growth performances and leaf biomass production in different sites of Sicily and in other regions of southern Italy and on the characterization and use of leaf powder for the obtainment of functional foods with high nutraceutical value.

Furthermore, the promotion of Moringa cultivation aligns with Sicily's rich agricultural heritage and can foster community engagement and education. Workshops and training sessions can be organized to teach farmers about the benefits of Moringa, its cultivation techniques, and potential market opportunities. This not only empowers local communities but also strengthens the agricultural sector as a whole.

In summary, Moringa offers a multifaceted solution to the challenges posed by climate change in Sicily. By embracing this resilient crop, Sicilian farmers can enhance their income, improve soil health, and contribute to a more sustainable agricultural future. As the world looks for innovative ways to adapt to climate change, Moringa stands out as a beacon of hope for both farmers and the environment.

TABLE 4 Total mineral nutrient content in *M. oleifera* leaves at the end of each experimental year (2021–2022), planted using seeding (S) and transplanted (T) at two distances (50 and 100 cm).

Treatment	Ca	K	Mg	Na	Fe
2021					
kg ha^{-1}					
S50	26.9 Ab	10.4 Ab	4.9 Ab	2.4 Ab	0.6 Ab
S100	25.6 Ab	10.9 Ab	3.8 Bb	1.8 Ab	0.3 Bb
T50	149.0 Aa	43.8 Aa	23.9 Aa	7.5 Aa	3.5 Aa
T100	103.6 Ba	42.9 Aa	14.1 Ba	5.6 Aa	1.2 Aa
2022					
kg ha^{-1}					
T50	67.4 A	69.6 A	12.2 A	1.6 A	0.4 A
T100	44.7 B	45.2 B	7.1 B	0.6 A	0.2 B

Different capital letters indicate significant differences ($p < 0.05$) between the two distances (50 and 100 cm) within the same planting mode (S or T). Different lowercase letters indicate significant differences ($p < 0.05$) between the two planting modes (S or T) at the same distance (50 and 100 cm). Reported results are expressed as mean ± standard deviation of three field replicates.

Data availability statement

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

Author contributions

GS: Data curation, Formal analysis, Methodology, Validation, Writing – original draft. CG: Conceptualization, Methodology, Resources, Supervision, Visualization, Writing – original draft, Writing – review & editing. VL: Data curation, Formal analysis, Validation, Writing – original draft. CL: Formal analysis, Writing – original draft. SM: Formal analysis, Writing – original draft. GG: Formal analysis, Writing – review & editing. SO: Visualization, Writing – original draft. GF: Project administration, Resources, Writing – review & editing. MM: Conceptualization, Funding acquisition, Project administration, Resources, Writing – original draft.

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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/fsufs.2025.1576147/full#supplementary-material

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