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Effects of alternative protein sources from food industry byproducts in starter feeds on dairy calf growth performance under different conditions: a preliminary study

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Introduction: This study evaluated the effects of alternative protein sources from food industry byproducts in starter feeds on dairy calf growth and metabolism under different feeding conditions.

Methods: Sixty Holstein × Black-and-White female calves were allocated into six groups across two commercial farms. Farm 1 included T1 (Starter A: 15% egg powder), T2 (Starter B: 15% feed yeast), and CON1 (standard starter); Farm 2 included T3, T4 (identical to T1 and T2, respectively), and CON2 (standard starter). All groups were monitored over a 90-day period for feed intake, weight gain, feed conversion ratio (FCR), and blood biochemical parameters. Starters also included milk whey and Bacillus subtilis as a probiotic. Statistical analyses were performed using ANOVA (P \leq 0.05 considered significant).

Results: At Farm 1, calves in group T2 demonstrated significantly greater weight gain (+3.47%, P = 0.01) and improved FCR (P = 0.03) compared to the CON1 group. Additionally, T2 calves showed elevated total protein (P = 0.04) and alkaline phosphatase (ALP) activity (P < 0.05), suggesting enhanced metabolic efficiency. At Farm 2, while growth and FCR differences were not statistically significant (P > 0.05), calves in T3 showed increased total protein at day 60 (P = 0.04) and elevated gamma-glutamyl transferase (GGT) at day 30 (P = 0.02).

Discussion: These preliminary findings suggest that incorporating food industry byproducts in starter feeds may improve feed efficiency and

metabolic parameters in dairy calves. However, their effectiveness appears to depend on feeding conditions, including basal diet composition and feed palatability. Further investigation is needed to clarify these interactions and assess long-term effects.

KEYWORDS

sustainable animal husbandry, dairy calves, nutrition, egg powder, feed yeast, milk whey, growth performance, feed conversion

1 Introduction

Food waste is a market failure causing over USD 1 trillion in losses annually, with 13% of global food reserves lost from harvest to retail (Programme, 2024). The FAO general director emphasized abandoning the linear "get-produce-throw away" model and transforming agrifood systems to be more efficient, inclusive, resilient, and sustainable (International Day of Awareness of Food Loss and Waste: FAO calls for circular model in agrifood systems, n.d). Food waste, generated across production, processing, and supply chains, is rich in nutrients such as carbohydrates, proteins, lipids, and minerals (Conrad and Blackstone, 2021). In terms of mitigating micronutrient loss, exacerbated by environmental degradation, food waste remains one of the most underutilized resources (Diaconeasa et al., 2022; Dou et al., 2024). Addressing food waste reduces its environmental impact, with repurposing as animal feed being an effective solution (Nath et al., 2023). Advanced research is needed to convert food waste into cost-effective feed and other valuable products (Cederberg and Sonesson, 2011; Aleisa and Alsaleh, 2024; Sun et al., 2024; Wang et al., 2024).

Globally, unprocessed food waste has been used successfully, particularly in the context of swine and poultry production (Boumans et al., 2022). In Kazakhstan, its use was common during the development of pig farming but has since declined, and its use in ruminants is limited by the risk of infectious dis-eases due to inadequate processing (Ominski et al., 2021). In our study the Kostanay region of Kazakhstan alone generates significant volumes of food industry by-products with strong potential for conversion into animal feed. Daily poultry egg losses reach up to 6,000 units at individual enterprises, while dairy facilities discard more than 48,000 kg of whey. In industrial egg production, a portion of the output is rejected due to cracks, shell defects, structural abnormalities, or contamination. Such eggs lose their nutritional value and, according to international studies, are unsuitable for sale or processing, resulting in direct production losses (Alig et al., 2023). Furthermore, a single distillery produces up to 240 tons of spent grains annually. These figures demonstrate that even at the regional level, valuable organic resources remain largely unutilized. Similar patterns can be observed across Kazakhstan, suggesting that the country as a whole possesses substantial reserves of food industry waste suitable for feed production.

To address these challenges, the high nutritional value of food industry by-products, such as egg powder, whey, and nutritional yeast, offers a promising opportunity for their conversion into valuable feed ingredients. Egg powder contains approximately 48.2g of protein, 43.5g of fat, and 5.5g of carbohydrates per 100 grams, providing around 599 kcal (Abreha et al., 2021). Whey protein offers about 65.6g of protein, 4.5g of fat, and 7.2g of carbohydrates per 100 grams, contributing to 398 kcal (De Wit, 1998). Nutritional yeast provides 50g of protein, 6.5g of fat, and 30g of carbohydrates per 100 grams, with a total energy content of 350 kcal (Patterson et al., 2023). Given the high nutritional value of such raw materials, we decided to explore the possibility of processing and using them in calf feed. Raising dairy calves presents both economic and physiological challenges for the dairy industry. It is also worth noting that many farmers face the challenge of choosing feed options (Belli et al., 2024). It is noted that the use of food waste as animal feed could contribute to a more circular and sustainable food production. Innovative approaches are being pursued to improve the well-being, gastrointestinal development, and productivity of calves (Dame-Korevaar et al., 2021).

Currently, researchers are increasingly focusing on insects as a feed ingredient, with insects such as black soldier fly larvae, mealworms, and crickets representing a sustainable alternative protein source for animal feed, offering high nutritional value and a significantly lower environmental footprint compared to conventional feed ingredients (Fu et al., 2025). However, in Kazakhstan, the use of insects as a protein source for cattle has not gained widespread adoption due to limited research in this area. A Kazakhstan-based study by (Kuchar et al., 2025) investigated the use of black soldier fly (*Hermetia illucens*) larvae as a feed ingredient. The authors highlighted the high nutritional value of the formulated feed.

High-quality animal-based protein is particularly important for calves, due to their underdeveloped rumen limiting digestion during early growth stages. Increased protein levels in calf diets positively influence their growth dynamics (Ockenden et al., 2023), and protein quality remains a key factor in their development (Brosh et al., 2000; Raeth et al., 2016). Researchers have investigated alternative nutrient sources for calves, focusing on their potential to enhance growth and health through the provision of high-grade protein content (Kim et al., 2022; Thornton et al., 2023). While starter feed is crucial for calf growth, findings on specific components remain mixed.

To validate the effectiveness of alternative protein sources, this study developed and tested starters based on food production waste to evaluate the effectiveness of this approach. The study was conducted using Black-and-White x Holstein calves as these breeds are the leading dairy breeds in Kazakhstan, where they dominate commercial milk production. The hypothesis of the study was that animal protein could be as effective as plant-based components in the composition of starter feeds for ruminant animals. The objectives of study were to develop starters from food production waste, including defective eggs from poultry farms, feed yeast derived from distillery stillage, and milk whey, and to assess their nutritional value, safety, and impact on the growth of Holstein x Black-and-White calves during the milk-feeding period.

Our research demonstrated that the aforementioned food production waste is safe and effective for dairy calves, emphasizing the importance of low-waste production in raising strong and healthy livestock.

2 Materials and methods

2.1 Formulation, nutritional characteristics and microbial purity of experimental calf starters

The experimental starters were formulated based on the recommendations of NRC (Committee on Nutrient Requirements of Dairy Cattle, 2021) and designated as follows: starter A contained 15% egg powder, which was derived from broken or damaged eggs at the production facility that were unsuitable for human consumption. Starter B was formulated using 15% feed yeast as the primary component. Both starters were based on 20% rapeseed meal, 33% ground yellow corn, and 20% ground barley, with 10% dried milk whey powder added to enhance the taste of the starters. Additionally, both formulations included 1% "Calfostonic" premix and 1% "Vetom 1.1" probiotic which contained live spore-forming bacteria Bacillus subtilis (B. subtilis). The technology for producing egg powder included cleaning eggs from shells, mixing egg whites and yolks, pasteurization at 65°C, and drying using the spray dryer SD-8000G (Labfreez Instruments, Changsha, Hunan, China). The inlet temperature of the spray dryer was 180°C, and the outlet temperature was 80°C. Feed yeast was produced by fermenting a substrate (distiller's grain) using a Saccharomyces cerevisiae (S. cerevisiae) culture, and was provided by LLP "Bioenergy Kazakhstan." Dry whey was produced through whey concentration, and spray drying. The drying temperature regime was 175°C at the inlet and 75°C at the outlet. All these components (after preliminary dosing) were mixed using a horizontal feed mixer, followed by granulation using a pellet machine with a matrix cell size of Ø4 mm. The composition and nutrient levels of the experimental starters presented in the Table 1.

The chemical composition of the experimental starters as well as the basal diet of calves was determined using various methods, including the measurement of dry matter (DM) and moisture content by drying samples at 105°C for 6 hours according to ISO 6496:1999 "Feed. Moisture definitions" (ISO 6496:1999 "Feed. Moisture definitions," 1999), the determination of ash content via TABLE 1 Ingredients and chemical composition of the starters fed to calves.

Ingredients, % by weight	Starter A	Starter B
Ground yellow corn	33.0	33.0
Ground barley	20.0	20.0
Rapeseed meal	20.0	20.0
Egg powder	15.0	-
Feed yeast	-	15.0
Milk whey powder	10.0	10.0
"Calfostonic" premix ¹	1.0	1.0
"Vetom 1.1" probiotic ²	1.0	1.0
Total	100.0	100.0
Chemical composition		
Dry matter (%)	89.26	89.33
Crude protein (%)	20.58	20.20
Fat (%)	7.09	2.04
Crude fiber (%)	3.40	3.49
Ash (%)	3.17	3.11
Nitrogen free extract (%)	55.03	60.49

¹Contained per 1 kg: A 600,000 IU; Vitamin D3 200,000 IU; Vitamin B1–100 mg; Vitamin B2–200 mg; Vitamin B6–10 mg; Vitamin B12 1.1 mg; Vitamin E 75 mg; Vitamin K3–25 mg; Nicotinic acid 1.25 g; Calcium pantothenate 500 mg; Choline chloride 25.0 g; DL-methionine 10.0 g; L-lysine 2.5 g; Gentian root 5.0 g; Carnitine hydrochloride 3.0 g; Sodium glutamate 7.5 g; Sodium chloride 25.0 g; Magnesium carbonate 5.0 g; Sodium selenite 36.5 mg; Manganese (as sulfate) 480.0 mg; Cinc (as sulfate) 504.0 mg; Iron (as sulfate) 600.0 mg; Copper (as sulfate) 125.0 mg; Cobalt (as sulfate) 105.0 mg; Potassium iodide 124.0 mg; Calcium carbonate 443.9 g; Dicalcium phosphate 300.0 g.

²Contained per 1 g: 1×10⁶ CFU of live microbial cells of the *B. subtilis*.

burning samples in a muffle furnace following ISO 5984:2022 "Animal feeding stuffs - Determination of crude ash" (ISO 5984:2022 "Animal Feeding Stuffs - Determination of Crude Ash," 2022), the analysis of crude protein (CP) using the Kjeldahl method on the KjeltecTM 8400, (FOSS, Denmark) analyzer as per ISO 5983-1:2005 "Animal feeding stuffs - Determination of nitrogen content and calculation of CP content" (ISO 5983-1:2005 "Animal Feeding Stuffs - Determination of Nitrogen Content and Calculation of Crude Protein Content," 2005), the measurement of fat using the Soxhlet extraction method on the SOXTECTM 8000 (FOSS, Denmark) extractor in accordance with ISO 6492:1999 "Methods for the determination of crude fat" (ISO 6492:1999 "Methods for the Determination of Crude Fat," 1999), and the determination of crude fiber (CF) via near-infrared spectroscopy using the FOSS NIRSTM 2500D (FOSS, Denmark) analyzer as per ISO 12099-2017 "Feed, grain and products of its processing" (ISO 12099:2017 "Feed, grain and products of its processing", 2017). Milk analysis was performed using MilkoscanTM FT2 (FOSS, Denmark) according to ISO 21543:2020 "Milk and milk products - Guidelines for the application of near infrared spectrometry" (ISO 21543:2020 "Milk and milk products - Guidelines for the application of near infrared spectrometry", 2020). The content of nitrogen-free extracts (NFE) was calculated

by the difference method according to Nutritional Ecology of the Ruminant (Soest, 1994), subtracting the main components of the feed from the total dry matter:

$$\%$$
NFE = $\%$ DM - ($\%$ CP + $\%$ EE + $\%$ CF + $\%$ Ash)

where: NFE = nitrogen free extract, DM = dry matter, EE = ether extract or crude fat, CP = crude protein, CF = crude fiber.

The determination of the microbiological purity of starters was performed in accordance with ISO 7218:2024 "Microbiology of the food chain - General requirements and guidance for microbiological examinations" (ISO 7218:2024 "Microbiology of the food chain -General requirements and guidance for microbiological examinations," 2024). The standards for microbiological safety followed the Technical regulations of the Customs Union "On food safety (TR CU 021, 2011). In the microbiological analysis, a sterile peptone saline solution (0.9% NaCl) was employed, along with the following culture media: meat peptone agar (MPA) for assessing total bacterial count (TBC), Sabouraud agar for the total fungal count (TFC), Endo agar for detecting Escherichia coli (E. coli), and selenite broth and bismuth sulfide agar for the detection of Salmonella species. Incubation conditions varied for each sample: for TBC, incubation was carried out at 37°C for 48 hours; for TFC, at 25-28°C for 7 days; and, for E. coli and Salmonella, at 37°C for 24 hours. The colony-forming units (CFU) on the Petri dishes were counted using the following formula:

$$CFU/g = \frac{(N \text{ colonies } *N \text{ dilutions})}{Culture \text{ volume (ml)}}$$

All the tested starters were found to comply with the microbiological safety standards, as the TBC did not exceed the allowable limit of 5×10^5 CFU/g. TFC values were below the permissible level of 5×10^4 CFU/g. No *E. coli* or *Salmonella* were detected in the samples (Table 2).

2.2 Experimental design

The starter testing was conducted in parallel from March to May 2024 on two farms in the Kostanay region of Kazakhstan: JSC "Zarya" in Arkhipovka village ($53^{\circ}30'$ N, $64^{\circ}12'$ E; farm 1) and LLP "Saryagash" in Pereleski village ($52^{\circ}32'$ N, $62^{\circ}05'$ E; farm 2). A total of sixty newborn Holstein × Black-and-White female calves (n = 60,

TABLE 2	Microbiological	analysis	of	starters.	
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Indicator	Regulatory safety indicators ¹	Starter A	Starter B
TBC, CFU/g	<5×10 ⁵	1.2×10 ³	1.8×10 ³
TFC, CFU/g	$<5 \times 10^{4}$	2.0×10 ²	2.2×10 ²
E. coli, CFU/g	N/a	N/d	N/d
Salmonella, CFU/g	N/a	N/d	N/d

¹Technical regulations of the Customs Union "On food safety" (TR CU 021, 2011), TBC, total bacterial count; TFC, total fungal count; CFU, colony-forming units; N/a, Not allowed; N/d, Not detected.

30 per farm, average body weight 39.96 ± 0.23 kg) were allocated into six treatment groups using the group-analogs method. At farm 1, the groups consisted of T1 (Starter A, n = 10), T2 (Starter B, n = 10), and control group 1 (CON1, Viamin ViaCorn 40150 standard starter at farm 1, n = 10). An identical grouping was employed at farm 2, forming T3 (Starter A, n = 10), T4 (Starter B, n = 10), and control group 2 (CON2 with KR-TU1 standard starter at farm 2, n = 10).

After birth, all calves were separated from their mothers and fed colostrum (4.0 L/day/calf) for the first 72 hours. Until the age of 2 months, the calves from both farms were kept in individual pens $(150\times120 \text{ cm})$ with wheat straw bedding which was replaced every two days, and after reaching 2 months, they were moved to group housing, where each calf was provided with a space of 2.5 m². Disinfection in the calf housing was carried out every week. The total duration of the experiment was 90 days.

During the experiment, each calf from the test and control groups received water ad libitum and a basal diet, which is described below in Table 3. The calf diet varied depending on the month and farm. On farm 1, calves received whole milk (6.00 L/day) and starter feed (0.25 kg/day) during the first month. By contrast, farm 2 used an equivalent volume (6.00 L/day) of "Calvomilk" milk replacer, which was prepared by dissolving the powder in warm water (~38°C) at a 1:7 ratio. In the second month, both farms introduced hay (smooth bromegrass on Farm 1 and ryegrass on Farm 2) at 0.50 kg/day, along with silage (grass silage on Farm 1 and cereal silage on Farm 2) at 1.00 kg/day. The starter feed was increased to 0.50 kg/day. In the third month (61-90 days), milk and milk replacer were completely removed, while the diet was supplemented with 1.00 kg/day of starter feed and 0.50 kg/day of grain mix. The hay and silage continued to be provided, with their daily intake increasing to 1.00 kg and 1.50 kg, respectively. Different feed compositions were used on the two farms to evaluate the effectiveness of the starters under varied feeding conditions.

2.3 Study of growth dynamics

For weighing calves, The TCS-1500 digital scales (Shanghai Yousheng Weighing Apparatus Co., Ltd, China) were used with an accuracy of \pm 0.2 kg. BW gain was monitored through average daily gain (ADG), absolute gain (AG). ADG was calculated using the following formula, where W₁ — Final weight (kg); W₀ — Initial weight (kg); and T — the number of days:

$$ADG(g/d) = \frac{W_1 - W_0}{T}$$

AG, reflecting the difference in weight between age periods, was calculated as the difference between the final and initial weights for each period:

$$AG(kg) = W_1 - W_0$$

Body measurements, including height at the withers, trunk length and chest girth were taken with standard instruments such as measuring sticks and tapes. Regular weighing is crucial for monitoring calves' growth, as it reflects their health and condition (Silva et al., 2024).

TABLE 3 Basal diet of calves.

Distances		Periods				
Diet com	ponent	4–30	days	30–60 days		60–90 days
Farm 1						
Whole	Milk	6.00 I	_/day	6.00 L/day		-
Smooth brom	negrass hay	Adapt	ation	0.50 kg/day		1.00 kg/day
Grass s	ilage	Adaptation		1.00 k	g/day	1.50 kg/day
Grain 1	mix ¹	-		-	-	0.50 kg/day
Starte	ers	0.25 k	g/day	0.50 k	g/day	1.00 kg/day
Farm 2						
Milk rep	lacer ²	6.00 I	_/day	6.00 1	L/day	-
Ryegras	s hay	Adapt	ation	0.50 k	g/day	1.00 kg/day
Cereal s	silage	Adapt	ation	1.00 k	g/day	1.50 kg/day
Grain	mix	-		-	-	0.50 kg/day
Starte	ers	0.25 k	g/day	0.50 kg/day		1.00 kg/day
Chemical compos	Chemical composition ⁵					
Diet Component	DM%	CP%	Fat%	CF%	Ash%	NFE%
Farm 1				'		'
Whole milk	12.27	3.39	3.64	_	_	5.24
Smooth bromegrass hay	92.24	8.75	1.30	62.73	6.28	13.18
Grass silage	35.21	3.73	1.44	20.71	8.21	1.12
Grain mix	88.32	13.77	2.43	5.14	2.62	64.36
Control starter 1	89.30	20.67	2.04	9.47	6.08	51.04
Farm 2						
Milk replacer	95.06	18.95	17.32	_	10.63	48.16
Ryegrass hay	95.17	6.11	2.26	33.41	4.91	48.48
Cereal silage	39.25	14.13	1.65	5.28	6.75	11.44
Grain mix	88.32	13.77	2.43	5.14	2.62	64.36
Control starter 2	88.06	19.85	2.41	3.61	6.32	55.87
Experimental Starter	rs					
Starter A	89.26	20.58	7.09	3.40	3.17	55.03
Starter B	89.33	20.20	2.04	3.49	3.11	60.49

¹Grain mix contained per 1 kg: barley 30%, oats 10%, peas 10%, wheat 50%.

²⁴ Calvomilk" milk replacer diluted with water at a 1:7 ratio and contained per 1 kg: whey, whole dried milk, protein-fat concentrate, flax meal, vitamin-mineral complex, probiotic, flavoring. ³⁴ Viamin ViaCorn 40150" starter. Ingredients per 1 kg: extruded wheat, wheat, corn, sunflower cake, soybean meal, chalk, premix PK 5–1 Start.

⁴KR-TU1 starter. Ingredients per 1 kg: soft wheat, barley, corn, corn gluten, soybean meal, sunflower oil, beet molasses, dried whey, table salt, monocalcium phosphate, limestone flour. ⁵DM, dry matter; CP, crude protein; CF, crude fiber; NFE, nitrogen free extract.

2.4 Feed intake measurement and feed conversion ratio calculation

For the recording of feed consumption, each group was provided with separate rations in individual buckets for each calf.

Prior to feeding, each portion of feed was weighed daily using digital scales, with the initial mass recorded as (M_0) . At the end of the day, the remaining feed in the bucket was weighed again, with the final mass recorded as (M_1) . The actual daily feed consumption was calculated as the difference in dry matter intake (DMI) of the feed,

where M_0 — the initial DM weight of the feed (kg), M_1 — the remaining feed DM weight (kg):

$$DMI = M_0 - M_1$$

The feed conversion ratio (FCR) was determined for the entire experimental period (1–90 days) using the following formula, where DMI — dry matter intake (g/day) and ADWG — average daily weight gain (g/day):

$$FCR = \frac{DMI}{ADG}$$

2.5 Biochemical blood analysis of calves

Blood samples were collected from calves 24 hours after birth and at the end of every month from the jugular vein using 8 mL vacutainers with a clot activator. The samples were then centrifuged at 3,000 x g for 15 minutes, and the serum was harvested for biochemical analysis. The analysis was conducted using the BioChem FC-120 (High Technology, Inc., USA) biochemical analyzer, measuring total protein, alkaline phosphatase (ALP), glucose, non-esterified fatty acids (NEFA), urea and gammaglutamyl transferase (GGT). Regular monitoring of the physiological condition of animals is crucial in the context of northern Kazakhstan, given the high prevalence of diseases, in this line, Bermukhametov et al. emphasized the need for improved control and prevention strategies to mitigate the associated impacts on cattle health and productivity (Bermukhametov et al., 2024).

2.6 Statistical analysis

The statistical data analysis was conducted using SPSS software (IBM SPSS Statistics 27, Chicago, Illinois, USA). The normality of distribution was tested using the Kolmogorov-Smirnov test, and the homogeneity of variances was tested using Levene's test. For each farm, a one-way analysis of variance (ANOVA) was performed: on Farm 1, the T1, T2, and CON1 groups were compared, and on Farm 2, the T3, T4, and CON2 groups. Subsequently, Tukey's HSD test was applied for multiple comparisons within each farm. Statistical differences between groups were indicated by letter superscripts (a, b, c; A, B, C): groups sharing the same letter showed no significant differences, whereas groups with different letters were significantly different. Different letters indicate statistically significant values between groups, with lowercase letters (a, b, c) representing significance at $P \le 0.05$ and uppercase letters (A, B, C) indicating a higher level of significance at P < 0.01. The analysis included body weight, absolute gain, average daily gain, dry matter intake, feed conversion ratio, withers height, trunk length, chest girth, as well as blood biochemical parameters. The analysis was performed separately for each age of calves (1, 30, 60, and 90 days). The results are presented in tables as mean values and SEM (standard error of the mean), where SEM represents an estimate of the TABLE 4 Daily feed intake of dairy calves at farm 1 as a dry matter intake per day.

ltere	т	reatmen	t1	SEM ²	Р	
Item	T1	T2	CON1		- Value	
Feed intake from	4 to 30 d	days				
Starter DMI ³ (g/d)	208.72	207.23	207.53	1.29	0.49	
Milk DMI (g/d)	736.2	736.2	736.2	0.11	0.97	
Total DMI (g/d)	944.92	943.43	943.73	1.06	0.35	
Feed intake from 31 to 60 days						
Starter DMI (g/d)	438.6 ^a	437.57 ^a	417.69 ^b	2.67	0.02	
Milk DMI (g/d)	736.2	736.2	736.19	0.09	0.95	
Hay DMI (g/d)	422.69 ^b	440.28 ^a	439.96 ^a	2.85	0.04	
Silage DMI (g/d)	344.36	345.92	344.8	1.13	0.32	
Total DMI (g/d)	1,941.86	1,959.97	1,938.64	4.89	0.21	
Feed intake from	61 to 90	days				
Starter DMI (g/d)	878.96 ^a	871.36 ^a	832.54 ^b	2.78	0.01	
Hay DMI (g/d)	826.35 ^b	854.65 ^a	846.95 ^a	18.74	0.02	
Silage DMI (g/d)	519.13	520.68	518.97	2.08	0.39	
Grain mix DMI (g/d)	879.12	880.26	879.85	1.58	0.56	
Total DMI (g/d)	3,103.56 ^a	3,126.95 ^a	3,078.3 ^b	8.06	0.03	

 $^1\mathrm{Treatments}$: T1 — Starter A (15% egg powder; 10% milk whey powder) + Basal diet; T2 — Starter B (15% feed yeast; 10% milk whey powder) + Basal diet; CON1 — Viamin ViaCorn 40143 (Standard starter) + Basal diet. Different letters show statistically significant values between groups — a; b; c (P \leq 0.05).

²SEM, standard error of the mean.

³DMI, dry matter intake.

dispersion of the obtained means around the true population mean. The level of statistical significance was set at $P \le 0.05$.

3 Results

3.1 Experiment in the Farm 1

Analyzing feed intake, we found that from 31 to 60 days, starter DMI was significantly higher in T1 and T2 compared to CON1 (P = 0.02), with a 4.75% increase in T2. Hay DMI was also significantly higher in T2 and CON1 than in T1 (P = 0.04). From 61 to 90 days, starter DMI remained significantly higher in T1 and T2 than in CON1 (P = 0.01), with an increase of 4.66% in T2. Total DMI was also greater in T1 and T2 compared to CON1 (P = 0.03), averaging 0.82% and 1.58% higher in T1 and T2 respectively. In the same period, hay DMI was significantly greater in T2 than in T1 (P = 0.02). No significant differences were observed in milk, silage, or grain mix DMI (P > 0.05, Table 4).

Significant differences in body weight were observed at 90 days (P = 0.01), with calves in T2 averaging 108.89 kg (\pm 0.52), 3.47% higher than CON1 (105.23 kg \pm 0.52). Trunk length and chest girth in T2 were also greater than in CON1 by 1.28% and 3.91%,

TABLE 5 Growth performance and feed conversion ratio of dairy calves at the farm 1.

Daviana at ava	Treatment ¹			CEM2	
Parameters	T1	T2	CON1	SEM	P - value
Initial body weight (3 d, kg)	40.14	39.82	39.53	0.19	0.43
Final body weight (90 d, kg)	107.52 ^b	108.89 ^a	105.23 ^b	0.52	0.01
Initial height at the withers (3 d, cm)	77.60	77.73	78.09	0.42	0.95
Final height at the withers (90 d, cm)	89.86	90.03	90.19	0.16	0.23
Initial trunk length (3 d, cm)	70.28	70.06	70.36	0.11	0.54
Final trunk length (90 d, cm)	93.39 ^b	94.30 ^a	93.11 ^b	0.19	0.03
Initial chest girth (3 d, cm)	73.21	73.11	73.30	0.09	0.73
Final chest girth (90 d, cm)	106.27 ^b	109.50 ^a	105.39 ^b	0.22	0.01
Gains from 4 to 30 d					
Average weight gain (kg)	22.16 ^a	22.59 ^a	20.93 ^b	0.29	0.05
Average daily weight gain (g/d)	820.72 ^a	836.82 ^a	775.24 ^b	10.91	0.05
Height at the withers change (cm)	4.82	4.43	4.15	0.24	0.14
Trunk length change (cm)	8.12	8.27	8.09	0.13	0.09
Chest girth change (cm)	10.89 ^b	11.34 ^a	10.56 ^c	0.42	0.01
Feed conversion ratio	1.15	1.12	1.21	0.07	0.33
Gains from 31 to 60 d					
Average weight gain (kg)	23.65	24.55	23.74	0.30	0.39
Average daily weight gain (g/d)	788.27	818.30	791.48	9.67	0.39
Height at the withers change (cm)	1.81	1.89	1.92	0.56	0.23
Trunk length change (cm)	7.74 ^a	7.93 ^a	7.52 ^b	0.21	0.01
Chest girth change (cm)	11.55 ^a	12.06 ^b	11.27 ^a	1.24	0.01
Feed conversion ratio	2.46	2.39	2.44	0.03	0.25
Gains from 61 to 90 d					
Average weight gain (kg)	21.57	21.93	21.03	0.25	0.34
Average daily weight gain (g/d)	719.16	731.07	700.97	8.32	0.42
Height at the withers change (cm)	3.13 ^b	2.86 ^c	3.6 ^a	0.24	0.05
Trunk length change (cm)	7.26 ^b	8.04 ^a	7.14 ^b	0.12	0.02
Chest girth change (cm)	10.62 ^B	12.99 ^A	10.26 ^B	2.22	<0.01
Feed conversion ratio	4.32 ^b	4.27 ^b	4.39 ^a	0.08	0.02
Gains from 4 to 90 d					
Average weight gain (kg)	67.38 ^a	69.07 ^a	65.70 ^b	0.51	0.02
Average daily weight gain (g/d)	795.87 ^a	816.13 ^a	775.04 ^b	5.86	0.02
Height at the withers change (cm)	7.14	7.30	6.29	0.46	0.56
Trunk length change (cm)	23.12 ^b	24.24 ^a	22.75 ^b	1.09	0.01
Chest girth change (cm)	33.06 ^b	36.39 ^a	32.09 ^b	2.49	0.01
Feed conversion ratio	2.57 ^b	2.53 ^b	2.63 ^a	0.06	0.03

¹Treatments: T1 — Starter A (15% egg powder; 10% milk whey powder) + Basal diet; T2 — Starter B (15% feed yeast; 10% milk whey powder) + Basal diet; CON1 — Viamin ViaCorn 40143 (Standard starter) + Basal diet. Different letters show statistically significant values between groups — a; b; c ($P \le 0.05$) and A; B; C (P < 0.01). ²SEM, standard error of the mean (n = 10 for T1, T2 and CON1). respectively (P = 0.03, P = 0.01). Between 4 and 30 days, weight gain was significantly higher in T1 and T2 than in CON1 (P = 0.05), with T2 showing the largest difference of 1.66 kg (7.92%). Chest girth gain during this period was also significantly higher in T2 (P = 0.01). From 31 to 60 days, trunk length gain in T1 and T2 exceeded that of CON1 (P = 0.01), while chest girth gain was significantly higher in T2 (P = 0.01). Between 61 and 90 days, we observed significant differences in withers height gain, which averaged highest in CON1 (P = 0.05), while T2 averaged the highest trunk length (P = 0.02) and chest girth gains (P < 0.01). Over the full 90day period, weight gain in T2 was significantly higher than in CON1 (P = 0.02), with trunk length and chest girth increases of 1.49 cm (P = 0.01) and 4.3 cm (P = 0.01), respectively. Feed conversion ratio

 TABLE 6 Biochemical blood parameters of dairy calves at the farm 1.

was significantly lower in T1 and T2 compared to CON1 at 90 days (P = 0.03, Table 5).

In the biochemical blood analysis, total protein concentration was significantly higher in T2 (74.00 g/L) compared to CON1 (71.70 g/L) at 90 days (P = 0.04), with an increase of 3.21%. No significant differences were found at 1 or 60 days. ALP activity showed notable increases in T2 at 60 and 90 days (P = 0.02, P = 0.04), rising by 3.91% and 3.97%, respectively, compared to CON1. Glucose levels were significantly higher in T2 at 30 days (P = 0.02), reaching 112.69 mg/dL, a 4.79% increase over CON1. NEFA levels were markedly reduced in T1 and T2 at 30 and 60 days (p = 0.01), with the most pronounced decrease observed in T1 at 60 days, where levels dropped by 64.71% compared to CON1. Urea concentration

Treatment ¹			CEM2			
Parameters	Age, days	T1	T2	CON1	SEM	P - Value
	1	67.12	66.80	67.20	1.58	0.68
Total protein (g/L)	30	74.15 ^a	73.95 ^a	72.38 ^b	0.76	0.05
	60	72.98	73.05	72.19	0.51	0.17
	90	72.44 ^b	74.00 ^a	71.70 ^b	0.67	0.04
	1	384.20	371.10	359.60	5.54	0.35
4 T D ³ (TT (T))	30	274.55	270.26	271.69	3.39	0.21
ALP ³ (U/L)	60	158.96 ^b	162.34 ^a	156.23 ^b	4.63	0.02
	90	136.20 ^a	138.90 ^a	133.60 ^b	3.26	0.04
	1	114.48	112.59	114.09	0.32	0.44
Glucose (mg/dL)	30	102.98 ^c	112.69 ^a	107.54 ^b	4.54	0.02
	60	87.45	90.32	87.63	2.51	0.09
	90	82.78	84.63	82.56	2.47	0.16
	1	0.48	0.39	0.41	0.02	0.38
	30	0.15 ^b	0.22 ^b	0.32 ^a	0.04	0.01
NEFA [*] (mmol/L)	60	0.12 ^b	0.19 ^b	0.34 ^a	0.05	0.01
	90	0.16	0.21	0.25	0.02	0.14
	1	11.40	10.61	11.85	0.02	0.27
	30	25.54 ^a	24.89 ^a	23.35 ^b	0.36	0.04
Urea (mg/dL)	60	27.36 ^b	30.6 ^a	23.92 ^c	1.06	0.01
	90	23.89	23.24	22.06	0.29	0.19
	1	559.63	612.34	574.05	8.61	0.24
0.0m ⁵ (11/1)	30	44.98	47.03	42.67	0.69	0.09
GGT (U/L)	60	38.25	39.71	37.53	0.35	0.11
	90	28.09	29.74	28.65	0.27	0.15

¹Treatment: T1 — Starter A (15% egg powder; 10% milk whey powder) + Basal diet; T2 — Starter B (15% feed yeast; 10% milk whey powder) + Basal diet; CON1 — Viamin ViaCorn 40143 (Standard starter) + Basal diet. Different letters show statistically significant values between groups a; b; c (P ≤ 0.05).

²SEM, standard error of the mean (n=10 for T1, T2 and CON1).

³ALP, alkaline phosphatase.

⁴NEFA, Non-esterified fatty acids.

⁵GGT, gamma-glutamyl transferase.

TABLE 7 Analysis of dry matter intake in calves at the farm 2.

ltown	Tı	reatmen	ıts ¹	SEM ²	P -			
item	Т3	T4	CON2		Value			
Feed intake from 4 to 30 days								
Starter DMI (g/d)	201.63 ^b	198.41 ^b	213.56 ^a	9.19	0.04			
Milk DMI (g/d)	814.80	813.44	813.31	0.14	0.97			
Total DMI (g/d)	1016.43	1011.85	1026.87	4.52	0.09			
Feed intake from 30 to 60 days								
Starter DMI (g/d)	440.14	442.31	440.3	1.61	0.63			
Milk DMI (g/d)	814.7	814.8	814.7	0.08	0.89			
Hay DMI (g/d)	473.11	474.08	473.56	1.45	0.47			
Silage DMI (g/d)	391.23	390.84	391.25	1.03	0.59			
Total DMI (g/d)	2119.18	2122.03	2119.81	3.14	0.16			
Feed intake from	n 60 to 9	0 days						
Starter DMI (g/d)	878.36	882.36	880.54	3.84	0.32			
Hay DMI (g/d)	948.71	949.69	950.14	2.31	0.16			
Silage DMI (g/d)	586.2	586.24	585.87	1.08	0.75			
Grain mix DMI (g/d)	880.1	879.5	880.32	1.58	0.52			
Total DMI (g/d)	3293.37	3297.79	3296.87	2.66	0.21			

¹Treatment: T3 — Starter A (15% egg powder; 10% milk whey powder) + Basal diet; T4 — Starter B (15% feed yeast; 10% milk whey powder) + Basal diet; CON2 — KR-TU1 (Farm 2 standard starter) + Basal diet. Different letters show statistically significant values between groups a; b; c ($P \le 0.05$).

 2 SEM, standard error of the mean (n = 10 for T3, T4 and CON2).

increased significantly in T2 at 60 days (p = 0.01), showing a 27.90% rise over CON1, while at 30 days, both T1 and T2 exhibited higher urea levels than CON1 (p = 0.04), with increases of 9.39% and 6.60%, respectively. GGT activity remained stable across all groups with no significant differences at any time point (P > 0.05, Table 6).

3.2 Experiment in the Farm 2

Studying feed intake parameters on Farm 2, we detected that starter intake differed significantly (P = 0.04) between groups at 1–30 days, likely due to adaptation to the new starter type. However, no significant differences were observed in total DMI across periods (Table 7).

The results on Farm 2 contrast sharply with those on Farm 1. While starters influenced growth performance on Farm 1, no such effect was observed on Farm 2 (P > 0.05, Table 8).

Analyzing the biochemical blood parameters of calves at Farm 2, we observed a significant difference in total protein at 60 days, where T3 had a higher concentration (P = 0.04) compared to T4 and CON2. We also noted GGT activity at 30 days, which was also significantly higher in T3 compared to T4 and CON2 (P = 0.02). No other significant differences were found in total protein across the other time points (P > 0.05, Table 9).

4 Discussion

Analyzing growth parameters of calves at both farms, we concluded that the effectiveness of starters is highly dependent on feeding conditions. At Farm 1, T1 calves (Starter A) and T2 (Starter B) exhibited significantly higher starter feed intake than CON1 during days 31-60 (P = 0.02) and 61-90 (P = 0.01). Intake in T2 increased by 4.75% and 4.66%, respectively. Total DMI was also significantly higher in T1 and T2 by 1.58% (P = 0.03), while no significant differences were noted in milk, silage, or grain mix intake (P > 0.05). Growth analysis revealed that T2 calves had a final body weight of 108.89 kg, exceeding CON1 by 3.47% (P = 0.01). Trunk length and chest girth in T2 increased by 1.28% (P = 0.03) and 3.91% (P = 0.01), respectively. In contrast, at Farm 2, no significant differences were found in feed intake or growth parameters among groups (P > 0.05). This discrepancy may be attributed to differences in basal diet composition, feeding management, and the adaptation period required for experimental starters. These findings align with previous research indicating that immunized egg protein improved body weight at 42 and 56 days, with a tendency for increased weight post-weaning (63-70 days) (Kuijk et al., 2021). However, other studies have found no impact of liquid egg additives on growth parameters or serum protein levels, despite their positive effects on immune function (Tufan et al., 2020).

FCR is a key indicator of nutrient utilization efficiency, reflecting how effectively feed is converted into body mass. Lower FCR values indicate better feed efficiency, meaning less feed is required per unit of weight gain. It is worth noting that feed additives can also play a major role in increasing the FCR (Davis et al., 2022). In our study, we observed a significant reduction in FCR in T1 and T2 compared to CON1 at Farm 1 (P = 0.03), indicating improved nutrient utilization with the experimental starters. Notably, T2 exhibited the highest feed efficiency, consistent with its superior weight gain and feed intake. These findings suggest that incorporating alternative protein sources in starter diets enhanced digestive and metabolic efficiency. In contrast, at Farm 2, no significant differences in FCR were observed among treatment groups (P > 0.05).

However, it was noted that the incremental addition of spraydried whole egg (SDWE) negatively affected the performance of bull calves fed commercial milk replacer (CMR). While the avidin content in CMR was not measured, the inclusion of 1 mg/kg of supplemental biotin did not improve performance, suggesting that avidin-related biotin deficiency may not have been the primary limiting factor (Quigley, 2002). Indirectly, our results are supported by the study of Santoro et al., 2004, in which the authors concluded that calves fed colostrum and egg protein-based milk replacer performed similarly to those fed colostrum and milk proteinbased milk replacer, but the feed conversion efficiency was lower in calves receiving the egg protein-based replacer (Santoro et al., 2004). This may be due to the presence of enzyme inhibitors (avidin, which binds biotin) and antinutritional compounds that may reduce amino acid availability. In milk replacers, avidin in egg protein may have reduced biotin absorption, potentially affecting calf growth. In newborn calves, protease activity (pepsin, trypsin) is

TABLE 8 Growth performance and feed efficiency of calves at the farm 2.

Description		Treatments ¹	CEM2	D. Malara		
Parameters	Т3	Т4	CON2	SEM-	P - value	
Initial body weight (3, kg)	39.9	39.91	39.96	0.12	0.96	
Final body weight (90 d, kg)	109.43	108.81	109.89	0.35	0.45	
Initial height at the withers (3 d, cm)	76.42	77.05	77.51	0.47	0.66	
Final height at the withers (90 d, cm)	91.52	90.97	90.89	0.16	0.22	
Initial trunk length (3 d, cm)	70.01	70.09	70.12	0.12	0.93	
Final trunk length (90 d, cm)	95.24	96.30	95.73	0.59	0.19	
Initial chest girth (3 d, cm)	73.14	73.25	73.04	0.11	0.74	
Final chest girth (90 d, cm)	111.18	109.87	110.69	0.38	0.14	
Gains from 4 to 30 d						
Average weight gain (kg)	22.20	22.11	22.18	1.41	0.98	
Average daily weight gain (g/d)	822.39	818.75	821.34	3.91	0.94	
Height at the withers change (cm)	5.14	4.87	5.02	0.21	0.12	
Trunk length change (cm)	7.93	8.27	7.87	0.12	0.24	
Chest girth change (cm)	11.98	11.67	11.82	0.14	0.18	
Feed conversion ratio	1.23	1.24	1.25	0.02	0.58	
Gains from 31 to 60 d						
Average weight gain (kg)	25.58	25.93	25.55	1.29	0.77	
Average daily weight gain (g/d)	852.71	864.44	851.69	7.83	0.74	
Height at the withers change (cm)	5.07	4.57	4.19	0.09	0.16	
Trunk length change (cm)	8.41	8.83	8.63	0.13	0.36	
Chest girth change (cm)	12.89	12.21	12.59	0.25	0.59	
Feed conversion ratio	2.47	2.46	2.47	0.02	0.51	
Gains from 61 to 90 d						
Average weight gain (kg)	22.18	22.21	21.19	0.44	0.47	
Average daily weight gain (g/d)	725.76	695.29	740.07	14.83	0.48	
Height at the withers change (cm)	4.89	4.48	4.17	0.22	0.35	
Trunk length change (cm)	8.89	9.11	9.08	0.16	0.74	
Chest girth change (cm)	13.17	12.74	13.24	0.16	0.38	
Feed conversion ratio	4.45	4.46	4.47	0.04	0.43	
Gains from 4 to 90 d						
Average weight gain (kg)	69.56	68.90	69.93	1.77	0.43	
Average daily weight gain (g/d)	772.88	765.54	776.98	3.60	0.44	
Height at the withers change (cm)	15.10	13.92	13.38	0.21	0.22	
Trunk length change (cm)	25.23	26.21	25.58	0.29	0.34	
Chest girth change (cm)	38.04	36.62	37.65	0.42	0.27	
Feed conversion ratio	2.66	2.65	2.66	0.02	0.32	

¹Treatment: T3 — Starter A (15% egg powder; 10% milk whey powder) + Basal diet; T4 — Starter B (15% feed yeast; 10% milk whey powder) + Basal diet; CON2 — KR-TU1 (Farm 2 standard starter) + Basal diet. Different letters show statistically significant values between groups a; b; c (P \leq 0.05). ²SEM, standard error of the mean (n=10 for T3, T4 and CON2).

TABLE 9 Biochemical blood parameters of calves at the farm 2.

Development and Anna Idavia			Treatment ¹	CEM2		
Parameters	Age, days	Т3	Т4	CON2	SEM-	P - value
	1	52.50	54.20	51.43	0.44	0.32
	30	62.10	61.80	59.70	0.41	0.25
l otal protein (g/L)	60	65.77 ^a	59.05 ^b	57.92 ^b	1.34	0.04
	90	64.37	63.80	63.55	0.13	0.37
	1	422.70	405.53	396.29	4.24	0.79
$AID^3(II/I)$	30	198.17	209.63	211.25	2.25	0.18
ALP (U/L)	60	148.52	147.23	150.08	0.45	0.21
	90	134.47	140.62	131.47	1.48	0.09
	1	99.48	102.21	104.10	0.73	0.65
Chasses (mg/dL)	30	91.44	92.71	92.27	0.20	0.15
Giucose (mg/uL)	60	82.80	83.42	82.13	0.37	0.29
	90	80.14	81.03	80.47	0.26	0.56
	1	0.51	0.57	0.49	0.02	0.85
NEE Λ^4 (mm al/I)	30	0.21	0.20	0.23	0.01	0.45
NEFA (mmoi/L)	60	0.14	0.19	0.18	0.01	0.36
	90	0.11	0.18	0.13	0.01	0.19
	1	13.90	14.61	14.85	0.29	0.91
	30	21.27	22.90	19.85	0.88	0.74
Urea (mg/dL)	60	25.14	27.36	28.10	0.79	0.35
	90	23.22	24.04	23.57	0.24	0.68
	1	421.32	408.41	439.97	9.16	0.89
$CCT^{5}(UU)$	30	52.15 ^a	38.54 ^b	40.30^{b}	4.27	0.02
GG1 (U/L)	60	36.21	38.88	36.85	0.80	0.29
	90	24.48	25.22	21.11	1.26	0.11

¹Treatment: T3 — Starter A (15% egg powder; 10% milk whey powder) + Basal diet; T4 — Starter B (15% feed yeast; 10% milk whey powder) + Basal diet; CON2 — KR-TU1 (Farm 2 standard starter) + Basal diet.

Different letters show statistically significant values between groups a; b; c (P \leq 0.05).

²SEM, standard error of the mean (n=10 for T1, T2 and CON1).

³ALP, alkaline phosphatase.

⁴NEFA, Non-esterified fatty acids.

⁵GGT, gamma-glutamyl transferase.

not yet fully developed, which may limit the breakdown of egg protein. Milk protein, especially casein and whey proteins, is broken down and absorbed more rapidly (Wood, 2022), whereas egg protein is digested more slowly, which could potentially limit its effectiveness. We assume that the bioavailability of egg powder in our study may have been higher due to reduced effects of antinutritional factors. In our study, we did not directly measure avidin levels, but we hypothesize that the thermal processing (pasteurization at 65°C and spray drying) may have partially inactivated avidin, thus minimizing its adverse impact on calf growth. According to the literature, avidin begins to lose its secondary structure at a temperature of approximately 75 °C. Complete denaturation occurs when heated to 100 °C for 10 minutes, and full inactivation is achieved at 121 °C after 25 minutes (Singh and Ramaswamy, 2014; Dhakal et al., 2020).

The increased feed intake was likely due to the improved palatability of the yeast-based starter, as the wheat-based yeast substrate created a sweeter taste, making it more attractive to calves. This is indirectly confirmed by the fact that various yeast-based products have been shown to improve productivity and health in calves. Although numerous studies have been conducted, the mechanism of action of *S. cerevisiae* in calves remains insufficiently understood (Alugongo et al., 2017). Feed yeast, included in T2 and T4, probably has been linked to improved immune responses and rumen development (Zhang et al., 2022). However, the study by (Huuskonen and Pesonen, 2015) did not

reveal a significant effect on average daily gain, although it was established that yeast additives are safe for animal consumption. Despite these inconsistencies, yeast is known to enhance oxidativereductive balance and intestinal health, improving immunity and stress resistance. In the study by (Maggiolino et al., 2023) dietary supplementation with a feed additive containing yeast products (yeast cell walls and hydrolyzed yeast) and microalgae contributed to the improvement of redox balance and gut morphology, enhancing the immune response of calves and their resistance to stress. In the long term, the implementation of feed yeast may also be effective for adult animals. This is confirmed by (Takiya et al., 2024), where the efficiency of nitrogen conversion into milk protein increased with the inclusion of yeast supplements, and the 3.5% fatcorrected milk yield (FCM) tended to increase in the yeast-fed groups compared to the control group. In the author's study, given that cows typically emit 250-500 L of methane per day, it was shown that two samples of spent brewer's yeast containing hop acids (humulones and lupulones) reduce methane production more effectively than the standard antibiotic monensin and can promote ruminant growth while simultaneously decreasing greenhouse gas emissions (Bryant et al., 2021). However, the impact of yeast on daily weight gain remains inconsistent, necessitating further research (Stefańska et al., 2018).

The role of milk whey in calf diets has been widely debated due to inconsistent research findings. Some studies suggest that whey can effectively replace milk replacers in supporting growth (Huuskonen, 2017), while others found no direct impact of dry whey in starters on weight gain (Lammers et al., 1998). Despite these contradictions, dry whey remains a valuable ingredient in calf nutrition due to its high content of lactose, bioactive proteins, and essential amino acids, which contribute to gut microbiota development, improved nutrient absorption, and enhanced immune responses. In the present study, no significant influence of whey-derived ingredients was observed, suggesting that starter composition as a whole plays a more critical role than individual ingredients alone. This aligns with findings that nutrient balance, ingredient interactions, and processing methods may affect the bioavailability and efficiency of whey components. Despite variations in reported effects, whey is recognized as a highly digestible energy source that promotes early rumen development and metabolic efficiency. These findings reinforce the notion that starter feeds are the most effective method for delivering essential nutrients during the intensive growth phase of calves, with their overall composition significantly influencing digestibility and growth performance (Ghaffari and Kertz, 2021; Eghtedari et al., 2024; Spina et al., 2024). Future research should focus on optimizing whey inclusion rates and evaluating its interactions with other dietary components to maximize its nutritional benefits and growth-promoting effects.

Examining biochemical blood parameters, we observed significant differences at Farm 1, where T2 (Starter B) calves exhibited a higher total protein concentration at day 90 (P = 0.04), with a 3.21% increase over CON1. ALP activity was also

elevated in T2 at days 60 and 90 (P = 0.02, P = 0.04). Glucose levels in T2 were significantly higher at day 30 (P = 0.02), suggesting improved energy metabolism. NEFA levels were lower in T1 (Starter A) and T2 (Starter B) at days 30 and 60 (P = 0.01), indicating enhanced lipid utilization. Urea concentration was significantly higher in T2 at day 60 (P = 0.01), reflecting differences in nitrogen metabolism. At Farm 2, the only significant differences observed were a higher total protein concentration in T3 (Starter A) at day 60 (P = 0.04) and increased GGT activity at day 30 (P = 0.02) compared to T4 (Starter B) and CON2. These findings suggest metabolic variations in response to different starter feeds but do not indicate a clear advantage in growth performance. The lack of significant differences in growth at Farm 2, despite variations in biochemical markers, suggests that external factors such as starter palatability and overall diet composition may have influenced feed intake and nutrient utilization In our previous study, it was established that a high total protein level in the blood indicates its active metabolism (Papusha et al., 2023). A notable aspect of the experimental starter formulations was the inclusion of B. subtilis, a probiotic microorganism known for its potential role in modulating gut microbiota, enhancing nutrient absorption, and supporting immune function in young ruminants. Previous studies have demonstrated that B. subtilis supplementation can enhance digestive enzyme activity, improve gut barrier function, and reduce inflammatory responses in calves. Additionally, its ability to produce bioactive compounds, such as proteases and antimicrobial peptides, may have supported gastrointestinal health, indirectly influencing overall metabolic efficiency (Davis et al., 2022).

5 Conclusions

This study demonstrated that incorporating food industry byproducts, such as egg powder and feed yeast, in starter feeds can enhance feed efficiency and metabolic parameters in dairy calves. However, the effectiveness of these alternative protein sources depended on feeding conditions, particularly the composition of the basal diet. While improvements in growth performance and biochemical markers were observed at Farm 1, no significant effects were detected at Farm 2, suggesting that external factors such as diet composition, feed palatability, and adaptation periods influence outcomes. Despite these promising results, several challenges remain. Variability in the nutritional quality of food byproducts, potential antinutritional factors in egg protein, and differences in rumen development among calves could impact digestibility and nutrient absorption. Additionally, ensuring consistent processing and microbial safety of byproduct-based feeds is crucial for their widespread application. Further research is needed to refine processing methods, optimize formulation strategies, and evaluate the long-term effects of these byproductbased starter components.

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.

Ethics statement

The animal study was approved by All procedures involving animal care and management were approved by the NLC Akhmet Baitursynuly Kostanay Regional University and Local Ethics Committee of the Research Institute of Applied Biotechnology (protocol number 3 approved 21 March 2024). The IRB registration number is IRB-00014274. The study was conducted in accordance with the local legislation and institutional requirements.

Author contributions

DM: Writing – review & editing, Visualization, Investigation, Conceptualization, Writing – original draft. BK: Data curation, Writing – original draft, Resources. MK: Data curation, Formal analysis, Writing – original draft. MS: Software, Writing – original draft, Investigation, Visualization. DN: Validation, Funding acquisition, Supervision, Project administration, Writing – original draft. JM: Writing – original draft, Project administration, Validation. NP: Writing – original draft, Funding acquisition, Supervision, Writing – review & editing, Project administration, Conceptualization, Methodology, Validation.

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