

EFFECTS OF INCREASING LEVELS OF AN ENERGY SUPPLEMENT ON PRODUCTIVE PERFORMANCE AND BEEF QUALITY OF FINISHING GRAZING STEERS

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ABSTRACT

The aim of the study was to assess the effects of increasing levels of an energy supplement on productive performance and the nutritional quality of steers finished on pasture. The supplement consisted of a blend of whole oat grain and steam-rolled corn. Treatments included a Control (steers grazing ryegrass pastures + mineral supplementation) and three increasing levels of the energy supplement corresponding to 0.5, 1.0, and 1.5% of body weight. Pasture nutritional quality was adequate throughout the experimental period, except for energy (< 2.88 MCal ME kg⁻¹ DM). Daily dry matter intake, body weight, and average daily gain were similar across treatments. However, carcass weight and dressing percentage differed, with lower values observed in the Control group. Steers receiving the 1.0% supplementation level exhibited greater fat cover and a tendency toward greater ribeye area. No treatment effects were observed in meat quality parameters. All samples were classified as tender (Warner-Bratzler Shear Force < 2.3 kgf), and pH was < 5.9. There were differences in the fatty acid profile, particularly in hexadecanoic and margaric acids, as well as in several C18:1 and trans fatty acid isomers. None of the treatments exceeded the recommended upper limit of 4 for the n-6: n-3 ratio. Overall, energy supplementation did not affect growth performance and beef quality; however, supplementation at 1.0% of body weight improved carcass weight and backfat thickness. This strategy may increase economic returns while maintaining a favorable fatty acid profile.

Key words: Fatty acids, omega-3, pastoral systems, sustainable beef.

INTRODUCTION

Beef-finishing grazing systems in temperate regions are limited to a small number of countries worldwide (Arias, 2024; Morales et al., 2015; Sales et al., 2019), but have gained increasing interest due to their potential for carbon sequestration (Khalil et al., 2020), provision of ecosystem services, and contributions to beef quality for human health (Arias et al., 2022; Nogoy et al., 2022; van Vliet et al., 2021). In these systems, pasture productivity and its nutritional quality are highly dependent on botanical composition and agroecological conditions (Duckett et al., 2013).

Grasslands in southern Chile are characterized by high pasture growth rates and superior forage quality during spring; however, productivity declines markedly during summer and winter (Doussoulin-Guzmán et al., 2022). This requires adjustments in the stocking rate to match the increased nutritional demands of animals during peak productive events (Mrad et al., 2009). It has been well established that the amount of forage produced determines the number of animals that can be maintained in a paddock, while forage quality determines the rate of animal growth and fat deposition. Therefore, periods of reduced pasture growth require the implementation of supplementation strategies in these systems (Arias, 2024). The productive performance and carcass quality in these systems are highly variable and influenced by several factors (Arias et al., 2019; Catrileo et al., 2014; Sales et al., 2019). The effects of finishing systems (grass-fed vs. grain-fed) on animal performance, carcass traits, and beef quality have been widely studied in Europe, Australia, and the USA, demonstrating that grass-fed beef has favorable sensory attributes and an advantageous fatty acid profile (Daley et al., 2010; Davis et al., 2022; Santos et al., 2021). However, these production systems differ substantially from those based on temperate grasslands found in South America and New Zealand.

Many producers supplement grazing animals with cereal grains to improve finishing performance and soundness. Research on feeding strategies for pasture-finished-steers consistently shows that targeted supplementation, combined with adequate pasture management, can substantially enhance productivity without fully transitioning to feedlot systems. There is a broad consensus that energy supplementation in grazing cattle improves rumen efficiency and animal performance, particularly under conditions of low forage quality, and that early-life supplementation may have residual benefits during the finishing phase (Caton and

Dhuyvetter, 1997; Naves et al., 2024). Similarly, Canozzi et al. (2023) found that moderate corn supplementation (around 0.6 - 0.8% of body weight; BW) in ryegrass-based systems significantly increased average daily gain (ADG) and carcass weight, highlighting the importance of balancing additive intake benefits against substitution effects at higher inclusion levels.

Complementary evidence from pasture-versus-feedlot comparisons indicates that pasture systems supplemented with concentrates can approach feedlot-level growth rates and carcass characteristics while maintaining grazing advantages, whereas pasture-only systems tend to have lower but still acceptable performance (Fruet et al., 2019). Furthermore, nutritional management during the finishing phase exerts a greater impact on final performance and carcass traits than earlier supplementation in the production cycle, reinforcing the strategic importance of late-stage nutrition (Simioni et al., 2021). Integrated management approaches combining supplementation with additives such as ionophores and growth-promoting technologies have been shown to reduce days to slaughter and improve overall system efficiency without compromising carcass quality (Fieser et al., 2007; Weiss et al., 2020). Collectively, these studies suggest that optimal performance in pasture-finished steers is achieved through moderate energy supplementation, strategic protein supply when forage quality declines, inclusion of high-quality legumes, and a strong emphasis on finishing-phase nutrition, rather than maximal concentrate feeding.

However, there is limited information regarding the effects of these strategies on productivity and beef quality under pasture-based systems. We hypothesized that the inclusion of cereal grains at 1% of body weight in grazing steers would improve productive performance and carcass quality without adversely affecting the fatty acid profile of beef. Accordingly, the present study aimed to assess the effects of increasing levels of energy supplementation on productive performance and nutritional quality of pasture-finished steers.

MATERIALS AND METHODS

All experimental procedures were conducted in accordance with animal welfare guidelines approved by the Agricultural Research Institute (INIA-Chile).

Animals and treatments

The experiment was carried out at INIA La Pampa Experimental Research Center

(40°51'24.88" S and 73°09'19.38" W). The study lasted 116 d, starting on November 16, 2018, and included a 30-day pre-experimental period for animal selection, castration, and adaptation to the production system. A total of 48 crossbred steers (Aberdeen Angus × Hereford), with an average age of 15 months (BW = 336 ± 25.6 kg), were used. Steers were randomly assigned to one of four treatments with increasing levels of energy supplementation (n = 12 per treatment). All groups grazed daily strips of the same pasture and received mineral supplementation (Veternal grazing, Metropolitan Region, Chile; 100 g/animal/day). The pasture consisted of a ryegrass cv. Herd (*Lolium x Boucheanum* Kunth) containing endophyte (AR1). Animals were slaughtered on March 12, 2019, at the Mafrius slaughter plant, located 48 km north of the research site. Final BW ranged from 473.7 to 501.3 kg depending on the level of energy supplementation.

Steers were randomly assigned to a control diet of pasture + mineral supplementation (CON) or to one of three treatment diets with increasing levels of energy supplementation expressed as a percentage of BW. Grazing was managed under a rotational system with a daily strip allocation. The treatment diets were defined as follows: CON + 0.5% BW (GS0.5); CON + 1.0% BW (GS1.0); and CON + 1.5% BW (GS1.5). The energy supplement consisted of a blend of whole oat grain and steam-rolled corn (Tables 1 and 2). Supplementation levels were adjusted fortnightly based on individual BW. Energy supplementation and mineral salts were fed daily at 15:00 h. Apparent pasture intake was estimated as the difference between pre- and post-grazing herbage mass within each allocated daily strip, estimated using a forage measuring plate (Farm Works F200, Ag Hub, Feilding, New Zealand). Pasture and energy supplement samples were collected weekly, pooled to obtain a monthly composite sample. Proximate analysis and fatty acid profiles were conducted at the INIA Remehue Laboratories.

Beef quality

In vivo measurements of ribeye area, backfat thickness, and marbling were obtained by ultrasonography. The same variables, along with muscle pH, were also measured postmortem (72 hours after slaughter). All measurements were taken from the *Longissimus dorsi* (LD) muscle. Ultrasound evaluations were performed one week prior to slaughter (March 6, 2019) by a qualified technician using an ultrasound system (Esaote Pie-medical Aquila pro model, Genoa, Italy). Ribeye area and backfat thickness were measured between the 12th and 13th ribs using two images per animal. Marbling was estimated

from five images per animal collected between the 11th and 13th ribs, at approximately 3/4 of the LD muscle length from the spine. Additionally, hip fat thickness (P8) was measured at the intersection of the *Gluteus medius* (rump) and *Biceps femoris* muscles in the hip region, parallel to the spine, using two images per animal. All images were processed using the same ultrasound system. Additionally, samples of the LD muscle (left side) were preserved for further instrumental analyses, including pH, color, texture, and fatty acid profile determination. Proximate analysis was carried out in duplicate on 50 g of ground, lyophilized beef samples from the LD. Muscle color was estimated using two 3.0 cm-thick LD steaks, which were thawed at 4 °C for a minimum of 48 h. The steaks were exposed to room-temperature for 30 to 60 min. Color was measured on the cut surface using a colorimeter (Konica Minolta, model CR-400, Tokyo, Japan), following Ruedt et al. (2023). Muscle pH was measured by taking two readings (cranial and caudal) of the LD muscle and using a pH meter (Hanna®, model 99163, Rhode Island, USA) and calculating their average. Texture was determined using the same steaks employed for color evaluation. The steaks were wrapped in aluminum foil and cooked in an electric oven (EKA, model KF 620, Camposampiero, Italy) until the internal temperature reached 71 °C. After cooking, samples were allowed to cool to room temperature for 30 min and subsequently stored at 4.0 °C ± 2.0°C for a minimum of 24 h. At 96 h post cooking, 6 to 10 subsamples (1.3 cm in diameter) were extracted and analyzed using a texture analyzer (Stable Micro Systems, PLUS-UPGRADE model, Surrey, England), equipped with a Warner-Bratzler shear force (WBSF) device. Shear force (kgf) and shear area (cm²) were recorded.

Fatty acid profile

The fatty acid profile of forage samples was analyzed using a combined method of direct transesterification and thin-layer gas chromatography, as described by Alves et al. (2008). Lipids from beef samples were extracted from 1.0 g of lyophilized muscle using a chloroform-methanol mixture (1:1, v v⁻¹), following Kramer et al. (1998). Lipid aliquots (~10 mg) from each steak were subjected to methylation using both acidic (methanolic HCl) and basic (sodium methoxide) reagents to ensure complete methylation of all lipids and thus avoid isomerization of conjugated linoleic acids (CLA) (Kramer et al., 1998; Aldai et al., 2005). For quantitative purposes, 1.0 mL of an internal standard (1.0 mg mL⁻¹ 23:0 methyl ester; n-23-M; Nu-Chek Prep Inc., Elysian, Minnesota, USA)

was added prior methylation. Fatty acid methyl ester (FAMES) contents were expressed as mg per 100 g of fresh beef and as a percentage (%) of total quantified FAMES.

Analyses were performed using a gas chromatograph (GC) equipped with a flame ionization detector (GC2010 Plus; Shimadzu®, Kyoto, Japan). For the study, a 100 m SP-2560 column (Supelco, Bellefonte, Pennsylvania, USA) was used with two complementary GC temperature programs, with final oven temperatures of 175 °C and 150 °C. In addition, a 100 m SLB-IL111 ionic liquid column (Supelco, Bellefonte, PA, USA) (Delmonte et al., 2011) was used to confirm the identification of several intermediate fatty acids resulting from biohydrogenation, including isomers of long-chain fatty acids (LCFA) and others. For both columns, hydrogen was used as the carrier gas at a constant flow rate of 1.0 mL/min, and the injector and detector temperatures were maintained at 250 °C. For peak identification, two reference standards were used: individual FAMES (21:0, 23:0, and 26:0) and a mixture of CLAs (9c, 11t-/8t, 10c-/11c, 13t-/ 10t, 12c-/). 8c, 10c-/9c, 11c-/10c, 12c-/11c, 13c-/11t, 13t-/10t, 12t-/9t, 11t-/8t, 10t-/18:2 was also used, which was obtained from Nu-Chek Prep Inc. (Elysian, Minnesota, USA). Isomerized mixtures of linoleic (18:2 n-6) and linolenic (18:3 n-3) acids (Sigma-Aldrich) were used as standards, and branched-chain fatty acids (BCFA) were identified using a bacterial FAME mixture (Matreya, Pleasant Gap, Pennsylvania, USA). Many of the trans-18:1 and CLA isomers, along with other non-conjugated dienes not included in the standard mixtures, were identified on their retention times and elution orders reported in the literature (Kramer et al., 2008; Alves and Bessa, 2009; Gómez-Cortés et al., 2009; Rego et al., 2009; Alves and Bessa, 2014) and further confirmed using the FAME fractions obtained from Ag + -SPE cartridges (Kramer et al., 2008; Belaunzaran et al., 2017).

Statistical analysis

The study factor was the energy supplement strategy, evaluated at four levels, using a completely randomized design. The observational unit for the productive performance variables was each steer, and the group of steers served as the unit for evaluating pasture and energy supplement intake within each treatment. For beef proximal and sensory variables, the observational unit was a sample of the LD muscle from each steer. Data were analyzed using one-way ANOVA and repeated-measures ANOVA in JASP version 0.18.3, with a significance level of 5%. Prior to analysis, data were tested for normality

(Shapiro-Wilk test) and homoscedasticity (Levene's test). Extreme outliers were removed prior to the repeated-measures analysis of ADG. The statistical models used were $Y_i = m + t_i + \varepsilon_{ij}$; where Y_{ij} = response variable, m = population mean, t_i = treatment effect, and ε_{ij} = experimental error. When there were statistical differences, the means were compared using Tukey's test; and $Y_{ijk} = \mu + \alpha_i + \beta_j + (\alpha\beta)_{ij} + (dl)_{ij} + \varepsilon_{ijk}$, including treatment effects (α_i), time (β_j), and its interaction ($\alpha\beta)_{ij}$, with $(dl)_{ij}$ corresponding to the random effect associated with the subject and ε_{ijk} . When variables included repeated measurements per animal (ADG and live BW), data were tested for sphericity using Mauchly's test (W). When the assumption of sphericity was violated, the Greenhouse-Geisser correction was applied. Multiple comparisons were performed using the Holm test. In addition, a dose-response analysis was conducted to evaluate linear, quadratic, and cubic polynomials trends.

RESULTS AND DISCUSSION

Diet quality and intake

The nutritional quality of the pasture was adequate, with crude protein (CP) ranging from 17.24% in November to 13.37% in February, and high *in vitro* digestibility (74.37% to 88.27%, respectively). Neutral detergent fiber (NDF) values were lower than 48% except for January, while the metabolizable energy content ranged from 2.88 in the spring to 2.48 (Mcal kg⁻¹ DM) in summer. As expected, the nutritional quality of oats and corn remained relatively constant (Table 1). Fatty acid profile in the pasture showed that polyunsaturated fatty acids (PUFA) ranged between 49.10 to 70.06%, while n-3 fatty acids ranged between 21.15 to 61.28%. On the contrary, PUFA averaged 41.65% for oats and 30.35% for corn, whereas monounsaturated fatty acids (MUFA) averaged 41.71% in oats and 52.58% in corn. In addition, omega-6 fatty acids averaged 40.67 and 51.24%. in oats and corn, respectively (Table 2). Steers received all the supplements offered in each treatment during the study. However, steers in the control group had higher apparent dry matter intake (DMI) than those receiving GS1.5 (similar to GS0.5) and showed a tendency to be higher than GS1.0.

Productive performance, carcass and beef quality

Steers' performance and carcass weights by treatment are summarized in Table 3. There were no differences between groups for total BW or total ADG ($p > 0.05$). In addition, no effects of

Table 1. Nutritional quality of the feed consumed by the steers during the experiment.

		DM	CP	Div	ME	NDF	EE	“D”	NEL	N	NSSC
	Month	(%)	(%)	(%)	(Mcal kg ⁻¹)	(%)	(%)	(%)	(Mcal kg ⁻¹)	(%)	(%)
Grass	Nov	92.27	17.24	88.27	2.88	42.00	2.41	80.14	1.71	2.82	28.51
Grass	Dec	92.09	14.40	81.65	2.71	47.74	2.02	74.76	1.62	2.30	27.83
Grass	Jan	90.37	15.10	76.08	2.56	50.10	2.80	70.19	1.54	2.42	23.97
Grass	Feb	91.91	13.37	74.37	2.48	46.37	2.95	67.86	1.49	2.14	28.50
Oat	Nov	88.39	11.19	75.57	2.66	31.99	6.32	73.15	1.59	1.79	47.81
Oat	Dec	88.74	11.39	74.82	2.63	27.87	6.08	72.26	1.57	1.82	51.87
Oat	Jan	88.88	11.42	75.74	2.88	32.66	5.79	73.89	1.60	1.83	47.43
Oat	Feb	89.58	11.50	75.92	2.68	27.46	6.11	74.02	1.60	1.84	52.46
Corn	Nov	86.62	6.74	96.12	3.38	7.11	2.04	95.31	1.99	1.08	83.38
Corn	Dec	86.85	6.92	96.71	3.44	7.58	1.92	97.26	2.02	1.11	82.81
Corn	Jan	87.67	7.13	97.03	3.40	8.74	2.99	96.14	2.00	1.14	80.16
Corn	Feb	87.23	6.90	96.72	3.44	7.17	1.86	97.40	2.02	1.10	83.33

DM: dry matter content, CP: crude protein, Div.: *in vitro* digestibility, ME: metabolizable energy, NDF: neutral detergent fiber, EE: etheral extract, V “D”: “D” value, NEL: net energy from lactation, N: nitrogen, NSSC: non-structural soluble carbohydrates.

interaction for energy supplement and time on BW ($p = 0.130$) and ADG ($p = 0.361$) were observed. As expected, there was an effect of time on BW and partial ADG ($p < 0.001$), whereas no main effect of energy supplement was detected for these variables ($p = 0.363$ and $p = 0.616$, respectively). It should be noted that this type of supplementation may enhance beef production per hectare through increased carrying capacity. In the present study, supplemented cattle exhibited lower apparent DMI (15.52 vs 13.92 kg for the control and average of the supplemented treatments, respectively). However, the control group exceeded the GS1.5 group by 4.34 kg. This is also supported by the dose-response analysis that showed a linear effect ($p < 0.05$) on BWf, HCW, CCW, and a quadratic effect for dressing percentage ($p = 0.0063$), reaching the highest value at GS1.0. The only exception was in ADG ($p = 0.448$), which did not show linear or quadratic effects. The maximum ADG (\pm SEM) was observed in the first period (2.41 ± 0.278 kg d⁻¹), while the lowest value was observed in the fifth period (0.43 ± 0.177 kg d⁻¹). In addition, carcass weights (hot and cold) and dressing percentage were lower in the control group of steers. Steers consuming above 1.0% BW as an energy supplement showed greater fat cover and a tendency for greater ribeye area when determined by ultrasound (Table 4). No changes in instrumental quality parameters were observed due to treatments ($p > 0.05$). In general, the L^* values were above 35, with positive a^* and b^* values, indicating that the fat is quite light in color, tending toward white ($L^* > 64$). WBSF values were < 2.27 across all treatments; thus,

all beef was classified as tender. In addition, pH values were < 5.9 in all treatments.

Steers did not show differences in final BW or ADG, contrary to the results reported by Klee et al. (2011), which may be explained by the rations being formulated with equal levels of digestible energy. This did not allow for differential weight gains due to the high grain levels (Maki et al., 2019), as the rations had no difference in energy concentration (Chen et al., 2024). The higher carcass weights (cold and hot) and higher dressing percentages observed in steers fed GS1.0 and GS1.5 could be explained by the gut-filling effect of pasture intake (Smith et al., 2021) and by the effects of visceral organs and hide (Coyne et al., 2019).

On the other hand, in feedlot systems with high-concentrate finishing diets, the inclusion of forage tends to reduce DMI because of the slower eating rate; however, productive performance is not affected by forage sources at similar levels of NDF (Swanson et al., 2017) or physically effective NDF and undigestible NDF (Pereira et al., 2021). Santos-Silva et al. (2023) concluded that high forage diets (forage-to-concentrate ratio of 54: 46 on a DM basis) reduced carcass dressing percentage in young bulls and increased redness and yellowness of subcutaneous fat, although without compromising commercial value.

In the present study, steers receiving an energy supplement exhibited higher ultrasound ribeye area and fat cover values were observed in steers fed an energy supplement. These findings are consistent with previous studies, which have reported ribeye area values of 58.9 and 61.5 cm²

Table 2. Relative fatty acid composition of dietary ingredients (%).

	Grass			Oat			Corn					
	Nov	Dec	Jan	Feb	Nov	Dec	Jan	Feb	Nov	Dec	Jan	Feb
SFA	22.12	37.01	28.83	28.34	16.45	15.50	16.21	17.75	16.84	17.48	16.57	17.21
16:1	0.10	0.41	0.13	0.26	0.16	0.12	0.16	0.12	0.10	0.53	0.08	0.12
18:1	2.53	5.91	7.18	18.06	40.16	41.60	41.92	39.66	29.57	29.95	30.64	28.87
T cis MUFA	2.68	6.44	7.46	19.07	41.10	42.40	42.77	40.36	29.91	30.74	30.97	29.24
16:1 trans	2.82	2.59	2.33	1.15	0.00	0.00	0.00	0.00	0.14	0.26	0.07	0.06
T trans MUFA	2.82	2.59	2.33	1.15	0.00	0.00	0.00	0.00	0.14	0.26	0.07	0.06
MUFA	5.49	9.03	9.79	20.22	41.10	42.40	42.77	40.36	30.05	31.00	31.03	29.30
PUFA	70.06	51.81	59.76	49.10	42.23	42.01	40.89	41.71	52.97	51.45	52.39	53.49
18:2 n-6	8.78	16.28	16.56	27.95	41.09	41.05	39.91	40.65	51.59	50.16	51.27	51.94
18:3 n-3	61.28	35.53	43.20	21.15	1.15	0.96	0.98	1.06	1.38	1.29	1.13	1.55
n-6	8.78	16.28	16.56	27.95	41.09	41.05	39.91	40.65	51.59	50.16	51.27	51.94
n-3	61.28	35.53	43.20	21.15	1.15	0.96	0.98	1.06	1.38	1.29	1.13	1.55

SFA = saturated fatty acids, 16:1= palmitoleic acid, 18:1 = oleic acid, T trans MUFA = total monounsaturated fatty acids (trans configuration), MUFA = monounsaturated fatty acids, PUFA = polyunsaturated fatty acids, C18:2 n-6: linoleic acid, C18:3 n-3: linolenic acid, n-6: n-6 fatty acids, n-3: n-3 fatty acids.

in treatments with the lowest and highest levels of grain supplementation, respectively (Ferreira et al., 2023). Ribeye area is an important variable because it is a predictor of beef yield (Wolcott et al., 2001) and shows a moderate but significant association with the proportion of high-value commercial cuts (Atencio-Valladares et al., 2008). The higher ribeye area may be attributed

to the combined effects of increased availability of metabolic substrates, hormonal signaling, and cellular hypertrophy. Specifically, by activating the propionate-glucose-insulin axis (Huntington, 1997), along with the IGF-1 and satellite cell proliferation (Schillo et al., 1992). Another potential explanation is that Angus cells proliferate more rapidly in high-nutrient

Table 3. Body weight, average daily gain (ADG), and carcass yield of steers grazing with increasing levels of energy supplementation in the diet.

Variable	Treatments				p-value
	CON	GS0.5	GS1.0	GS1.5	
BWi, kg	325.1 ± 25.0	334.7 ± 25.9	338.9 ± 20.7	345.8 ± 28.6	0.249
BWf, kg	473.7 ± 25.6	481.8 ± 21.0	492.0 ± 36.4	501.3 ± 40.4	0.179
ADG, kg d ⁻¹	1.34 ± 0.356	1.33 ± 0.166	1.38 ± 0.222	1.40 ± 0.199	0.868
HCW, kg	232.3a ± 14.7	248.4ab ± 13.3	254.6b ± 18.1	259.1b ± 19.8	0.002
CCW, kg	226.8a ± 14.3	243.0ab ± 13.0	249.1b ± 17.7	253.5b ± 19.4	0.001
Dressing, %	49.04a ± 1.2	51.58b ± 1.8	51.77b ± 1.1	51.74b ± 1.9	0.001

Means with different letters within columns are statistically different ($P < 0.05$).

BWi = initial live body weight; BWf = final live body weight at the farm; ADG: average daily gain; HCW = hot carcass weight; CCW = cold carcass weight; CON = ryegrass + mineral supplementation; GS0.5 = CON + 0.50% of BW as energy supplement; GS1.0 = CON + 1.00% BW as energy supplement; and GS1.5 = CON + 1.50% BW as energy supplement.

Table 4. *In vivo* and postmortem carcass characteristics of steers grazing with increasing levels of energy supplementation.

Variables	Treatments				p-value
	CON	GS0.5	GS1.0	GS1.5	
<i>Postmortem</i>					
Fat cover*	1.0a	1.2ab	1.6bc	1.7c	0.001
pH	5.54	5.52	5.52	5.47	0.539
Intramuscular fat (%)	1.08	1.33	1.42	1.42	0.339
REA, cm ² (10th intercostal space)	59.43	59.16	65.79	61.99	0.319
Back fat thickness, cm	0.37	0.42	0.45	0.47	0.460
<i>In vivo</i> (ultrasound)					
Back fat thickness, cm	0.40	0.35	0.41	0.33	0.251
QIB, %	7.10	7.21	6.79	7.31	0.604
REAU, cm ² (12th intercostal space)	63.09a	70.49ab	68.77ab	73.29ab	0.062
P8, cm depth	0.40	0.40	0.38	0.38	0.766

Means with different letters within columns are statistically different ($P < 0.05$).

* Fat cover of the carcass is a visual appreciation in the Chilean Norm of carcass classification with three increasing levels (1, 2, or 3); REA = ribeye area; QIB = Marbling; P8 = Hip fat thickness. CON = Ryegrass + mineral supplementation; GS0.5 = CON + 0.50% of BW as energy supplement; GS1.0 = CON + 1.00% BW as energy supplement; and GS1.5 = CON + 1.50% BW as energy supplement.

conditions (Coles et al., 2015). Nevertheless, intramuscular fat (marbling) did not differ among treatments, although it was numerically higher in steers fed with energy supplement. However, Park et al. (2018) indicated that high-nutrient or high-energy diets are required to achieve marked increases in muscle marbling and, consequently, increased fat deposition.

The pH ranged from 5.60 to 5.71, consistent with previous reports (Moloney et al., 2022; Sullivan et al., 2024). No differences in beef color attributable to the treatments were observed. However, previous studies have reported higher

L^* values (>35) (Morales et al., 2015; Moloney et al., 2022) and positive a^* and b^* values (del Campo et al., 2008; Pordomingo et al., 2013a; Pordomingo et al., 2013b). In the present study, fat color was lighter across all treatments, with L^* values >60, which is consistent with young animals fed high-energy diets (Pordomingo et al., 2013c; Santos-Silva et al., 2023). All steaks were classified as very tender, with no differences observed among treatments. Other researchers have reported similar results in beef cattle in Ireland (Moloney and Drennan, 2013), dairy biotypes in Chile (Morales et al., 2013; Catrileo et

al., 2014), and feedlot steers in Uruguay (Vignale, 2015). These values are expected for young, castrated animals with a high nutritional level and adequate aging time (three weeks; Bruce and Roy, 2019). In young animals, collagen solubility remains high until 14 months of age (Florek et al., 2022). Florek et al. (2022) indicate that cattle fed low-energy diets grow and mature more slowly than those fed high-energy diets, and that at a given chronological age, forage-fed cattle are physiologically less mature than grain-fed counterparts. Other studies have reported that a high nutritional level leads to a higher rate of collagen synthesis, which is on average less stable and, consequently, resulting in a muscle with greater tenderness (Bruce and Roy, 2019). On the other hand, calpain activity should also be considered, as it facilitates improved beef aging (Teira et al., 2006).

Fatty acid profile

Partial GC chromatograms (with final oven temperatures of 175 °C and 150 °C, respectively) showed different beef fatty acids from 16:0 to 11c-18:1 as well as their isomers. Significant differences were observed in the fatty acid profile, particularly for saturated fatty acids (SFA), including hexadecenoic (16:0) and margaric (15:0) acids ($p < 0.05$), which showed an inverse relationship with gain inclusion in the diet (Table 5). MUFA, particularly several 18:1 isomers, were higher in supplemented steers, whereas BCFA were lower in this group. PUFA did not present differences, except for 16:1, 9t, which showed an inverse relationship to the inclusion of grains, and 18:1, 10t, which increased with the incorporation of grains in the diet. No effects of supplementation were observed for EPA and DHA. Trans fatty acids in their different isomers showed differences according to the energy supplementation level ($p < 0.05$), with concentrations increasing as the level of grains in the diet increased. In contrast, vaccenic acid showed an inverse relationship, decreasing with increasing grain level. None of the treatments exceeded the upper limit of 4 for the n-6: n-3 fatty acid ratio established by the British Department of Health (Simopoulos, 2016). The ratios were 2.68, 2.37, 2.06, and 1.99 mg FA/100g fresh beef for GS1.5^τ, GS1.0^υ, GS0.5^ψ, and Control^ϑ, respectively ($p = 0.008$; similar superscript symbols indicate the same groups).

Beef has great nutritional importance given the high quality of its proteins and bioactive compounds, which contain all the essential amino acids, as well as its highly bioavailable minerals and vitamins (Pighin et al., 2016; Arias et al., 2022; Leroy et al., 2023). Likewise, fat, although

present in small proportions, is an important source of fatty acids, such as linoleic and linolenic acids, which are vital for maintaining the body homeostasis and for the synthesis of other essential acids, such as DHA and EPA (Swanson et al., 2012; Leroy et al., 2023). It is important to note that these properties were not affected by the treatments assessed, and the mean values fall within the range considered lean beef (An et al., 2019). Nevertheless, Whang et al. (2019) indicated that dietary energy level can influence ruminal microbiota and, in turn, rumen fermentation and fatty acid synthesis, showing high correlations between rumen bacteria and fermentation parameters and the intramuscular fat (IMF) fatty acid profile.

The relative distribution of the trans-18:1 isomer in ruminant fats will depend mainly on the basal diet fed. In forage-fed animals, t11-18:1 (vaccenic acid) would increase, whereas in high-digestible-starch diets, t10-18:1, will further increase (Aldai et al., 2013). In the present study, t6-8-18:1, t9-18:1, and t10-18:1 increased when steers were fed with GS1.5. The t10-18:1 is one of the main contributors of the total trans-18:1 content in partially hydrogenated vegetable oils, which correlates with cardiovascular disease (CVD) risk (Mozaffarian et al., 2009). In ruminants, t10-18:1 is associated with intensive feeding conditions and may become the major trans-18:1 isomer (Aldai et al., 2010; Aldai et al., 2013). In this context, to limit t10-18:1 in beef fat, grain supplementation should be restricted to 1.0% of BW.

Many studies have shown that a forage-based diet improves the muscle fatty acid profile from a human health perspective. In fact, it has been proposed that grass-fed beef could exert protective effects against several diseases, ranging from cancer to cardiovascular diseases, as evidenced by the increased functional n-3 fatty acids and decreased undesirable SFA (Davis et al., 2022; Nogoy et al., 2022). In addition, pastures have a higher proportion of PUFA (Najar-Villarreal et al., 2019; Davis et al., 2022), which aligns with the results obtained in the present study. Likewise, all treatments had a low n-6: n-3 ratio in intramuscular fat, ranging from 1.9 (CON) to 2.7 (GS1.5). These values remain below the maximum limit of 4.0 declared by the WHO (2003).

The studied strategy results in higher feeding costs; consequently, producers should consider the prices of available feedstuffs when formulating the energy supplement and evaluate the cost-benefit of its implementation. In addition, no growth-promoting implants were used in this study; under such conditions, fat deposition and fatty acid profiles could also be affected,

Table 5. Relative fatty acid composition (%) of beef from grazing steers consuming increasing levels of energy supplementation.

Fatty acids	Treatments				p-value
	CON	GS0.5	GS1.0	GS1.5	
<i>SFA (Saturated fatty acids)</i>					
10:0 (Ac. Capric)	0.06	0.05	0.05	0.06	0.389
12:0 (Ac. Lauric)	0.06	0.06	0.06	0.06	0.877
14:0 (Ac. Myristic)	2.50	2.44	2.48	2.41	0.952
15:0 (Ac. Pentadecanoic)	0.36a	0.32ab	0.29ab	0.26b	0.037
16:0 (Ac. Palmitic)	25.85	25.20	25.27	25.34	0.670
17:0 (Ac. Margaric)	0.90a	0.80ab	0.76ab	0.71b	0.025
18:0 (Ac. Stearic)	15.03	15.04	15.58	15.33	0.932
19:0 (Ac. Nonadecanoic)	0.08	0.07	0.06	0.06	0.140
20:0 (Ac. Arachidic)	0.09	0.09	0.10	0.08	0.362
22:0 (Ac. Behenic)	0.16	0.15	0.12	0.15	0.222
24:0 (Ac. Lignoceric)	0.03ab	0.04a	0.02b	0.03ab	0.051
<i>MUFA (Monounsaturated fatty acids)</i>					
9c-14:1	0.48	0.55	0.51	0.51	0.845
9t-16:1	0.10	0.07	0.06	0.08	0.147
10t-16:1	0.01	0.02	0.02	0.02	0.444
11t-12t-16:1	0.06	0.06	0.05	0.07	0.324
7c-16:1	0.22	0.21	0.21	0.21	0.724
9c-16:1	3.07	3.06	3.01	3.12	0.984
11c-16:1	0.09	0.11	0.11	0.11	0.789
14t-12c-16:1	0.03	0.03	0.03	0.03	0.898
13c-16:1	0.04	0.05	0.05	0.04	0.987
9c-17:1	0.62	0.56	0.53	0.53	0.084
6t-8t-18:1	0.11b	0.13ab	0.14ab	0.15a	0.022
9t-18:1	0.14b	0.16ab	0.18a	0.18a	0.010
10t-18:1	0.14b	0.18ab	0.22ab	0.28a	0.004
11t-18:1	1.15	0.96	0.91	0.89	0.191
12t-18:1	0.23b	0.26ab	0.28a	0.26ab	0.073
6c-8c-18:1	0.38	0.44	0.48	0.40	0.052
14t-18:1	0.01	0.02	0.01	0.01	0.865
15t-18:1	0.15	0.12	0.14	0.11	0.278
11c-18:1	1.12	1.14	1.13	1.21	0.544
12c-18:1	0.07b	0.10ab	0.12a	0.10ab	0.009
13c-18:1	0.20	0.24	0.23	0.24	0.413
15c-18:1	0.09	0.10	0.12	0.09	0.221
16c-18:1	0.05b	0.06ab	0.07a	0.06ab	0.011
16t-18:1	0.19	0.21	0.22	0.18	0.343
8c-20:1	0.06	0.06	0.05	0.04	0.041
11c-20:1	0.10	0.11	0.12	0.12	0.553
<i>PUFA (Polyunsaturated fatty acids)</i>					
8t,13c-18:2	0.08b	0.10a	0.10a	0.09ab	0.026
9t,12c-18:2	0.02	0.01	0.02	0.02	0.165
11t,15c-18:2	0.21	0.18	0.16	0.15	0.109
9c,15c-18:2	0.08	0.09	0.09	0.09	0.372
9c,11t-18:2	0.30	0.28	0.27	0.27	0.765
10t,12c-18:2	0.03	0.03	0.03	0.03	0.709
18:2 n-6	2.45	2.88	2.72	3.02	0.317
18:3 n-6	0.04	0.04	0.04	0.04	0.833

18:3 n-3	0.71	0.79	0.62	0.58	0.150
20:3 n-6	0.24	0.26	0.25	0.28	0.150
20:4 n-6	0.02	0.03	0.02	0.02	0.091
20:5 n-3 (EPA)	0.53	0.51	0.42	0.45	0.619
22:6 n-3 (DHA)	0.08	0.10	0.08	0.08	0.769
n-6	3.83	4.28	4.09	4.59	0.507
n-3	2.08	2.15	1.76	1.83	0.569
TOTALS					
16:1 cis	4.43	3.42	3.37	3.48	0.984
18:1 cis	35.50	36.17	36.66	36.11	0.714
16:1 trans	0.21	0.19	0.17	0.21	0.453
18:1 trans	2.47	2.47	2.56	2.45	0.956
cis MUFA	40.19	40.87	41.24	40.79	0.840
trans MUFA	2.68	2.66	2.73	2.66	0.985
TFA	6.01	5.96	6.07	5.90	0.986
CLA	0.36	0.34	0.32	0.33	0.703
SFA	46.69	45.65	46.10	45.64	0.765
MUFA	42.87	43.53	43.97	43.44	0.794
PUFA	5.96	6.49	5.89	6.47	0.800
HUFA	2.77	2.79	2.51	2.84	0.867
BCFA	1.68a	1.50ab	1.41ab	1.29b	0.041

Means with different letters within columns are statistically different ($P < 0.05$).

CON = ryegrass + mineral supplementation; GS0.5 = CON + 0.50% of body weight (BW) as energy supplement; GS1.0 = CON + 1.00% BW as energy supplement; and GS1.5 = CON + 1.50% of BW as energy supplement; TFA = trans fatty acids; CLA = conjugated linoleic acid; SFA = saturated fatty acids; MUFA = monounsaturated fatty acids; PUFA = polyunsaturated fatty acids; HUFA = highly unsaturated fatty acids; and BCFA = branched-chain fatty acids.

potentially yielding results different from those presented herein.

CONCLUSIONS

The results of this study suggest that the inclusion of energy supplementation up to 1.0% body weight did not affect the performance or beef quality of grass-finished steers, while improving carcass weight and backfat thickness. In addition, this strategy maintained a favorable n-6: n-3 fatty acid ratio. Therefore, it represents an effective feeding strategy to enhance animal performance and potentially improve economic returns for producers, while enabling compliance with industry standards for fat cover, fat color, and carcass conformation.

Author contributions

Conceptualization and Methodology, Rodrigo Morales. Software, Rodrigo Morales and Rodrigo Arias. Validation, Merbis Tesorero, Rodrigo Morales and Rodrigo Arias. Formal analysis, Merbis Tesorero, Rodrigo Arias. Investigation, Merbis Tesorero and Rodrigo Morales. Resources, Rodrigo Morales. Data curation, Merbis Tesorero

and Rodrigo Arias. Writing-original draft, Merbis Tesorero and Rodrigo Arias. Writing-review and editing, Merbis Tesorero, Rodrigo Morales, Ignacio Subiabre and Rodrigo Arias. Visualization, Rodrigo Morales. Supervision, Rodrigo Morales. Project administration, Rodrigo Morales. Funding acquisition Rodrigo Morales. All co-authors reviewed the final version and approved the manuscript before submission.

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