

Effects of the inclusion of brown seaweed (*Macrocystis pyrifera*) additive in the diet of grass-fed steers on carcass performance, meat quality, and nutrient composition

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ABSTRACT. The objective of this study was to evaluate the inclusion of a brown seaweed additive (SWA; *Macrocystis pyrifera*) in the diet of grass-fed steers on carcass performance, beef quality, and nutrient composition. A total of 20 Holstein-Friesian steers were randomly distributed into two groups: Control group (a basal diet without supplementation of SWA) and SWA group (2%-SWA) with basal diet + 30 g/day/animal of SWA during the breeding phase (11 months) and 48 g/day/animal of SWA during the fattening phase (4 months). Steers fed with 2%-SWA were not different ($P>0.05$) in final body weight, carcass weight, carcass dressing, fat thickness, ribeye area, and marbling score than those from the Control group. Likewise, no effects of 2%-SWA supplementation were detected ($P>0.05$) for beef quality traits, glycolytic potential, or their metabolites (muscular glycogen, glycose+glucose-6-phosphate, and lactate), evaluated in *longissimus lumborum* (LL) samples. Sensory evaluation showed a slight preference for Control group samples rather than those from the 2%-SWA group (58.93% and 41.07%; $P=0.06$). Regarding proximal composition, the inclusion of SWA only affected the total lipids present in the LL samples, which decreased significantly ($P=0.01$) in LL samples of grass-fed steers fed with 2%-SWA. The composition of macro (Ca, Na, Mg, P, and K) and micro (Mn, Fe, Cu, and Zn) minerals in LL samples were not affected ($P>0.05$) by the inclusion of SWA in the diet. The inclusion of the additive based on brown seaweed had not a detrimental effect on carcass performance, beef quality, and mineral content, however, it reduced the total lipids content in the LL muscle.

Keywords: seaweed, carcass, lipids, proximal composition, mineral, beef.

INTRODUCTION

Beef is known as one of the main sources of protein with high biological value, bioavailable minerals (Fe, Zn, and P), vitamins of the B-complex (B1, B2, B3, B6, and B12), and other nutritional components like vitamins D, E, and β -carotenes (Williams, 2007; Klurfeld, 2018). Besides, it is considered that meat has been related to the evolution of humanity due to its important impact on human cognitive, morphophysiological, and social development (Psouni *et al.*, 2012).

At present, beef consumption in Chile is estimated around 24 kg/year per capita, foreseeing a slight upward trend in the next 10 years (OECD-FAO, 2019). In addition, in the last decades, there is an increasing preference for natural, organic, and antibiotic-free meat (Karmaus & Jones, 2021), creating a growing interest in the study of natural supplements in animal nutrition. Also, beef industries are facing constantly challenges, such as an enhanced request for certification on animal welfare (Rossi *et al.*, 2020), environmental emissions, and climate change (Halmemies *et al.*, 2018). These demands require animal productions

to be more sustainable, in conjunction with the use of innovative resources (Halmemies *et al.*, 2018; Rossi *et al.*, 2020; Raja *et al.*, 2022). In this context, marine algae could be able to become an economical and competitive option for animal production worldwide (Raja *et al.*, 2022; Madeira *et al.*, 2017) due to their nutritional value, and content of bioactive compounds which could contribute to enhance production and health in animals (Halmemies *et al.*, 2018). Also, some seaweed species have shown the potential to mitigate ruminal methane production *in vitro* (Maia *et al.*, 2016).

Nutrition is one of the main factors that greatly influence growth and carcass performance (Guerrero *et al.*, 2013; Mwangi *et al.*, 2019), but it also produces important changes in the nutrient composition of meat, such as fatty acid composition, and mineral content (Rotta *et al.*, 2009).

Algae are autotrophic organisms with a simple structure, little or no cell differentiation and complex tissues, being considered talophytes. By taxonomy, they are classified into three groups: Chlorophyta or chlorophytes, Phaeophyta or pheophytes, and Rhodophyta or rhodophytes, which correspond to green algae, brown algae, and red algae, respectively (Quitral *et al.*, 2012).

Among the huge variety of marine algae, brown seaweed is widely found in large populations in North America (from Alaska to Baja California) and South America (from Perú to Tierra del Fuego), including the extensive Chilean coast (Baca *et al.*, 2008). Its large size and easily harvesting have allowed this type of algae to be studied and used in animal feeding (Makkar *et al.*, 2016). Within this group of algae, the best known in south America is *Macrocystis pyrifera* (popularly known as “huiró”), *Lessonia nigrescens*

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(“huairo negro”), and *Durvillaea antarctica* (“cochayuyo”) (Quitral *et al.*, 2012).

Brown seaweeds have a protein content of 5 to 15%, but very high levels of minerals, fatty acids, carbohydrates, and essential amino acids (Raja *et al.*, 2022; MacArtain *et al.*, 2007; Baca *et al.*, 2008; Makkar *et al.*, 2016). Particularly, *Macrocystis pyrifera* contains from 8.7 to 10.7% of crude protein, while the concentration of ashes ranged between 33.5 and 36.6% (Baca *et al.*, 2008). They are an excellent source of vitamins A, B1, B2, B3, B5, B12, C, D, E, and folic acid (Makkar *et al.*, 2016; Quitral *et al.*, 2012; Ortiz *et al.*, 2008). In addition, no anti-nutrients have been found in its composition (Casas *et al.*, 2005). In this context, brown seaweed could be considered a natural source of nutrients and bioactive compounds with great biological activity with potential benefits for animal health and growth, also as a choice to produce functional foods (Quitral *et al.*, 2012).

Extensive reviews about the inclusion of seaweeds in monogastric production have been published (Corino *et al.*, 2019; Angell *et al.*, 2016; Øverland *et al.*, 2019). However, fewer studies have investigated the effects of dietary seaweed on beef carcass traits, and its nutrient composition. The magnitude of the associate response to the inclusion of seaweed in the animals’ diet on growth performance, carcass traits, quality and nutrient composition of meat depends on the type of seaweed used, the bioactive components present in the extract, and the proportion and frequency used in the diet (Makkar *et al.*, 2016). The objective of this study was to assess the inclusion of a seaweed additive (*Macrocystis pyrifera*) supplied during the backgrounding and fattening phases of grass-fed beef cattle on its productive performance, carcass parameters, beef quality, and nutrient composition.

MATERIALS AND METHODS

ANIMALS AND SAMPLING

A total of 20 Holstein-Friesian calves, after weaning were reared, at then finished at the Agricultural Austral Research Station of the Universidad Austral de Chile. The calves, prior to the start of the trial, were managed in artificial rearing receiving milk replacer (4 L/day), initial concentrate (increasingly until reaching 2 kg/day), and alfalfa cubes, up to 200 g daily. The control and treatment groups were weaned averaging 79.6 and 77.8 days, weighing 98.0 kg and 99.4 kg, respectively. The backgrounding period lasted 11 months and the fattening period 4 months. Animals were fed mainly with permanent pasture (*Lolium perenne* L. dominated sward (55% *L. perenne*, 33% *Bromus valdivianus* Phil., 5% *Trifolium repens* L. and 7% of other species) offered based on 3% body weight measured at ground level. Animals were also supplemented with a commercial concentrate (49.3 maize, 11.5 soybean meal, 30.0 beet pulp, 4.6 beet

molasses), 4.5 of mineral mix and silage (approximately 17 kg/animal when needed) offered. The expected dry matter intake was 2.5% of body weight during the entire experiment. The pasture was managed on a rotational grazing with a resting period between 10 to 15 days. The concentrate and the silage were offered in feeding pens at 08:00 h and 15:00 h. Water was permanently offered *ad libitum* in the paddocks and enclosure pens. Samples of pasture, concentrate and SWA were collected on each period. All samples were immediately frozen at -20 °C and then freeze-dried for chemical analysis. Before chemical analysis, the samples were ground through a 1 mm screen (Willey Mill, 158 Arthur H, Thomas, Philadelphia, PA). The bromatological results of the analysis of forages, commercial concentrate, and SWA are described in Table 1.

Animals were assigned to one of the following treatments: Control group: basal diet with 0% of SWA; and 2%-SWA group: basal diet + 30 g/day/animal of SWA during the growing phase and 48 g/day/animal of SWA during the fattening phase. The levels of SWA used were determined in preliminary experiments. The additive was added to the concentrate and mixed manually and offered to animals in individual feeders. The SWA was fabricated by I+D Patagonia Biotecnología S.A., as an impalpable hydrolyzed seaweed powder produced by spray drying of *Macrocystis pyrifera*, that maintains its chemical-physical characteristics and bioactive compounds. Animals were sent to harvest when they reached between 16 to 18 months of age.

Table 1. Chemical composition of the experimental diet: forage, commercial concentrate, and seaweed additive (SWA).

Parameter	Forage	Commercial concentrate	SWA
DM, %	30.76	85.9	92.67
TA, %	7.15	5.81	25.60
CP, %	16.50	17.25	8.87
ADF, %	27.61	8.77	11.78
EE, %	0.29	3.41	0.29
CF, %	-	6.46	2.87
NDF, %	52.93	29.88	7.06
NFE, %	6.92	52.97	43.93
DV, %	73.31	77.83	70.91
ME, Mcal/kg DM	2.62	2.81	2.58

DM: Dry matter, TA: Total Ash, CP: Crude protein, ADF: Acid-detergent fiber Ether extract, and EE: Ether extract were determined by AOAC (1996). CF: Crude fiber determined by AOAC (1984). NDF: Neutral-detergent fiber obtained according to Van Soest *et al.* (1991). NFE: Nitrogen free extract. % NFE = 100 % - (% moisture + % EE + % CP + % TA + % CF). DV: Digestibility value according to Tilley & Terry (1963). ME: Metabolizable energy determined by Goering & Van Soest (1970).

CATTLE HARVESTING, CARCASS EVALUATION, AND SAMPLING

Steers were transported to a slaughterhouse plant facility located 15 km away from the farm and slaughtered after 12 h of lairage. Harvesting, dressing procedures, and *postmortem* inspection followed the standards of the Chilean regulation (INN, 1993). Final body (BW) and hot carcass weights were recorded to estimate carcass dressing yield. Ribeye area, fat thickness at the 10th rib, and degree of marbling were evaluated according to the procedure stipulated by the USDA (2017). Carcasses were chilled for 24 h *postmortem* at 2 °C.

A core of *longissimus lumborum* (LL) samples from each carcass was taken at 24 h *postmortem*, immediately frozen in liquid nitrogen (-196 °C) and stored at -80 °C for muscular glycogen content (MGC), glucose+glucose-6 phosphate (G+G6P), and lactate concentration (LC) determination. After 48 h *postmortem*, the entire portion of the LL muscle was removed from each left side of the carcass, and samples of 2.5 cm thickness were obtained. Two samples were used immediately for pH and color evaluation, and four samples of each loin were individually packaged and frozen at -20 °C during 30 days for the rest of the analysis.

MEAT QUALITY AND POSTMORTEM GLYCOLYTIC METABOLITES

A portable pH meter with a puncture electrode (Hanna, model HI 99163, Jud Cluj, Rumania) was previously calibrated with buffer pH 4 and 7 was used for pH measurement. A Hunter Lab Mini Scan XE Plus (Hunter Associates, Reston, VA, USA) was used with a 2.5-cm open port, Illuminant D65 and 10° standard observers to objectively evaluate color. Three readings were obtained from the muscle surface, and the mean was calculated. Readings were obtained after exposing the muscles to air for 30 min (bloom). The color scale used was Hunter L, a, b. The L value represents lightness; the a and b values represent redness, and yellowness, respectively.

The Warner-Bratzler Shear Force (WBSF) was estimated on samples cooked in a convection oven (Albin Trotter model E-EMB Digital) until reaching a final internal temperature of 70 °C following the guidelines of the American Meat Science Association (AMSA, 2016). The temperature was monitored using an oven thermometer ranged (-10 +110 °C; +/- 1°C) inserted into the geometric center of each steak. The cooked steaks were chilled for 2 h at 4 °C, and then eight cores (1.27 cm in diameter) were removed parallel to the muscle fiber orientation. Cores were sheared once each on the Warner- Bratzler Meat Shear apparatus (GR Manufacturing Co., Manhattan, KS, USA) to get WBSF values. The water holding capacity (WHC) was determined as cooking loss, which was determined by weight, expressed as a percentage compared to the

original weight of the sample. A taste preference test was performed in two sessions (56 panelists). Two steaks of each treatment were used in each session. The tests were carried out in individual evaluation cabinets illuminated with red light. Each panelist, in each session, tasted two samples (one from each treatment) at random, and they were asked to select the best preference.

The MGC, G+G6P, and LC were determined as described in Apaoblaza *et al.* (2015). Briefly, muscle samples were homogenized in ice-cold phosphate buffer (pH 7). Ten µL of homogenate were hydrolyzed in 200 µL of 0.1M HCl at 100 °C for 2 h, after which pH will be adjusted to 6.5-7.5 and glucose determined via NADP reduction with a linked assay involving hexokinase and glucose-6-phosphate dehydrogenase (Glucose HK 16-50 Sigma). The LC was determined from the homogenate via NAD reduction with a linked assay involving lactate dehydrogenase and glutamate pyruvate transaminase (Boehringer Mannheim). Glycolytic potential (GPOT) was calculated with the following formula $GPOT = (LC) + 2([MGC] + [G+G6P])$ and was expressed as millimoles of lactate per kilogram of muscle (Monin & Sellier, 1985).

NUTRIENT COMPOSITION OF BEEF

Moisture, protein, and lipid content of meat samples were determined according to the AOAC (1990). Duplicates of 10 g of ground meat were calcined in a furnace at 550 °C for 6 h. After cooling, the residue (white ash) was subjected to an acid digestion process with 10 mL of a 20% v/v hydrochloric acid solution by heating on a hot plate for 10 min. Mineral analyses were conducted by atomic absorption and/or atomic emission (AOAC, 1990), following the analytical methods described by Perkin-Elmer (1994). Values were expressed as g/100 g or mg/g of dry matter (DM).

STATISTICAL ANALYSIS

The experimental design was a completely randomized design. A one-way ANOVA was performed using a mixed model with SWA treatment as the main factor and animal as the random effect. The value $P \leq 0.05$ was used to declare the significant difference between the average scores. Tukey's multiple comparison test was used for the comparison of means. The Bonferroni correction was also performed to adjust the probability of P values. χ^2 test was used for sensory preference data. Analysis was performed using the R Program (R Core Team, 2021).

RESULTS

CARCASS PERFORMANCE.

Both groups had similar BW ($P=0.69$) at the beginning of the study with 463.9 kg and 470.6 kg for treatment and

control groups, respectively. Table 2 shows mean values for carcass traits. Carcass traits evaluated in this study were not affected ($P>0.05$) by the inclusion of 2%-SWA in the diet of steers. Most of the carcasses were described as practically devoid of marbling (scale 1; USDA, 2017), and all carcasses exhibited a similar fat thickness ($P>0.05$) and finish score or subcutaneous fat cover (1= Slight) according to INN (1993) and similar.

MEAT QUALITY TRAITS AND *POSTMORTEM* GLYCOLYTIC POTENTIAL AND THEIR METABOLITES

Table 3 shows the mean values for meat quality traits. There is no significant effect ($P>0.05$) of the inclusion of 2%-SWA in the diet of steers on muscular pH, cooking loss, and WBSF. Instrumental colour was determined in its three dimensions (a, b, and L values) and no significant differences were detected for any of them ($P>0.05$) when comparing both groups under study. The ANOVA detected the non-significant ($P>0.05$) effect of the inclusion of SWA on MGC, G+G6P, and LC and GPOT (Table 4) evaluated at 24 h *postmortem*.

Regarding sensory evaluation, in session 1, 18 panelists preferred samples from the control group equivalent to 64%. In session 2, it was counted 15 preferences for the control group (equivalent to 53%). Together, this represents 58.93%

Table 2. Effects of the inclusion of seaweed additive (SWA) in the diet of grass-fed steers on growth and carcass traits.

Variable	Control	2%-SWA	SEM	P-value
Final body weight, kg	470.6	463.9	8.10	0.69
Hot carcass weight, kg	227.88	221.36	4.49	0.48
Hot carcass dressing, %	48.37	47.70	0.28	0.24
Fat thickness, mm	4.27	4.06	0.29	0.73
Ribeye area, cm ²	45.61	49.93	2.32	0.37
Marbling*	1.8	1.6	0.17	0.59

* 1= practically devoid; 2= scarce; 3= small amount of marbling (USDA, 2017). SEM: standard error of the mean.

Table 3. Effects of the inclusion of seaweed additive (SWA) in the diet of grass-fed steers on meat quality traits.

Variable	Control	2%-SWA	SEM	P-value
Muscular pH, 48 h	5.65	5.67	0.03	0.69
Redness (a value)	15.21	15.52	0.29	0.62
Yellowness (b value)	9.72	9.62	0.25	0.74
Lightness (L value)	27.51	26.51	0.96	0.24
Cooking loss, %	16.41	16.44	0.64	0.96
WBSF, kg	2.19	2.12	0.09	0.46

SEM: standard error of the mean. WBSF: Warner Bratzler shear force.

Table 4. Effects of the inclusion of seaweed additive (SWA) in the diet of grass-fed steers on *postmortem* glycolytic metabolites and glycolytic potential.

Variable*	Control	2% SWA	SEM	P-value
MGC	6.07	6.73	0.69	0.64
LC	35.18	40.26	2.10	0.23
G+G6P	10.81	10.20	0.36	0.42
GPOT	65.63	59.56	3.90	0.45

SEM: standard error of the mean. MGC: muscular glycogen content. LC: lactate content. G+G6P: Glucose + Glucose-6-phosphate. GPOT: Glycolytic potential. *Measured at 24 h *postmortem* (mmol/kg).

of taste preference for Control samples compared to 41.07% of preference for 2%-SWA group. Although there was no statistically significant difference in the preferences of the panelists, a tendency ($P=0.06$) to prefer the samples of the Control group compared to those of the SWA group was observed. In both sessions, the panelists considered that all samples had a normal taste, without the presence of a strange or unpleasant flavor. Some panelists even highlighted the juiciness and tenderness of the samples in their observations.

NUTRIENT COMPOSITION OF MEAT

Table 5 shows the mean and standard error of the mean of the proximal composition and mineral content of LL, according to the treatment groups. Proteins and total ash were not affected by the SWA additive ($P>0.05$). However, there was an effect ($P<0.05$) of the inclusion of 2% of SWA on the total lipid content in the bovine LL. Samples from animals that were fed with 2% SWA had a lower ($P=0.01$; Table 5) amount of total lipids (5.24 g/100 g dry matter) than those from the Control group (6.65 g/100 g dry matter).

The inclusion of SWA in the diet of fattening steers did not affect ($P>0.05$) the content of the macro (Ca, Na, Mg, P, and K) and micro minerals (Mn, Fe, Cu, and Zn) evaluated in this study. Steers that were fed with 2% of SWA exhibited less numerical values of Mn and Zn than the Control group ($P>0.05$; Table 5).

DISCUSSION

In recent decades, interest in the use of seaweeds as organic ingredients in farm animal has increased (Makkar *et al.*, 2016). The inclusion of seaweed has been investigated in the feeding of sheep (Marín *et al.*, 2003), pigs (Baca *et al.*, 2008), rabbits (Rossi *et al.*, 2020), and cattle (Morrill *et al.*, 2017a, b). Most of these researchers have been focused on growth parameters, like body weight, daily weight gain and feed conversion. However, few studies of this nature have been conducted on the evaluation of carcass performance, meat quality, and nutrient composition.

Table 5. Effects of the inclusion of seaweed additive (SWA) in the diet of grass-fed steers on proximal composition and mineral content of *longissimus lumborum*.

Variable	Control	2%-SWA	SEM	P-value
Proximal composition ¹				
Ash	4.82	4.79	0.05	0.82
Crude protein	88.21	87.80	0.51	0.61
Total lipids	6.65	5.24	0.89	0.01
Macrominerals ¹				
Ca	0.017	0.017	0.001	0.98
Na	0.164	0.166	0.003	0.75
Mg	0.075	0.074	0.001	0.72
P	0.723	0.719	0.006	0.76
K	1.39	1.41	0.03	0.81
Microminerals ²				
Mn	8.82	7.97	0.764	0.59
Fe	26.07	26.71	2.58	0.90
Cu	15.22	15.62	0.83	0.81
Zn	77.29	72.68	2.75	0.42

¹ g/100 g of dry matter (DM). ² mg/g of DM. SEM: standard error of the mean.

In this study, the inclusion of 2% of SWA in the diet of grass-fed steers did not affect the final body weight. Several authors did not find significant variations in the growth of lambs (Al-Shorepy *et al.*, 2001) or steers (Anderson *et al.*, 2006) in response to the inclusion of supplements based on marine algae. On the other hand, Fike *et al.* (2001) reported an increase in the weight of lambs that were fed with seaweed extract during the summer grazing period. In Chile, only the studies of Mendoza (2017) and Nannig (2018) have evaluated the effect of brown algae on bovine, reporting similar average daily gains when comparing heifers treated with SWA vs. Control ones.

Carcass weight and dressing were not influenced by the inclusion of 2%-SWA in the diet of steers. Fat thickness, marbling, and ribeye area of the carcass from finishing steers fed with SWA were similar ($P > 0.05$) to those from the Control group. Morrill *et al.* (2017a) also reported no differences in carcass weight, fat thickness, or *longissimus* muscle area in carcasses from steers consuming 9% of post-extraction algal residue (PEAR) compared to those that received glucose infusion (Control group). However, in this same study, the marbling score was 15% greater in PEAR-fed group compared to Control carcasses.

The inclusion of 2% of SWA did not affect the instrumental tenderness of meat. Control samples of LL had 2.19 ± 0.09 kg in WBSF and the mean \pm SEM for the SWA group was 2.12 ± 0.12 kg. Jerez-Timaure *et al.* (2021) found similar results in pork LL samples. Morrill *et al.* (2017b) stated that feeding with PEAR resulted in a slight but no significant reduction of shear force, with values between 2.77 and 2.5 kg. Miller *et al.* (2001) developed

a tenderness threshold based on consumer acceptability, establishing that WBSF values < 3.0 kg can be classified as tender beef and WBSF values > 4.6 kg were tough beef. According to Miller *et al.* (2001), LL samples from this study could be categorized as tender (< 3 kg), and those are very similar to the values reported by Morrill *et al.* (2017b).

The sensory evaluation showed that a slight majority (58.93%) of panelists had a tendency ($P = 0.06$) to prefer samples of the Control group rather than those from the 2%-SWA. These results could be related to the increased content of total lipids detected in samples from the Control group (Table 5). Lipid compositions are related to flavor development, with a different range of flavor precursor being produced from saturated and polyunsaturated fatty acids (Wood & Enser, 1997). Braden *et al.* (2007) stated that the supplementation effect on sensory evaluation taste depends on the ingredient of the diet. Morrill *et al.* (2017b) found no significant differences, as in this study, in the tenderness and flavor of meat from bovines supplemented with an additive made of marine algae residues. Jerez-Timaure *et al.* (2021) reported similar results, they reported a greater taste preference ($P < 0.05$) by consumers in samples coming from the Control group. However, in the case of studies in rabbits, it is reported that supplementation with brown marine algae improved the palatability of their meats (Rossi *et al.*, 2020). Morrill *et al.* (2017b) stated that cattle have limited ability to digest and absorb lipids to negatively affect meat flavor. Also, it is well known that changes in fatty acid composition are often associated with flavor differences (Arshad *et al.*, 2018). In this study, the fatty acid composition was not evaluated. Jerez-Timaure

et al. (2021) did not detect differences in the fatty acid profile of loin samples from pigs that fed SWA at 2 and 4% compared to the Control ones (0%-SWA).

Meat color is considered one of the most valued and preferred attributes by the consumer at the time of purchase, preferring bright red meat and rejecting dull or brown meat (Holman *et al.*, 2017). It has been studied that supplementation with antioxidants, such as vitamin E, helps to extend the case and shelf life of fresh meat products, improving color stability, reducing lipid oxidation, and delaying the formation of metmyoglobin (Allen *et al.*, 2001; Braden *et al.*, 2007; Rossi *et al.*, 2020). It is important to emphasize that the additive based on brown marine algae *Macrocystis pyrifera* used in this study is rich in antioxidants (MacArtain *et al.*, 2007; Ortiz *et al.*, 2008), and an improvement in meat color was expected. However, instrumental color values of 2%-SWA samples were not statistically different ($P>0.05$) compared to the control ones. Jerez-Timaure *et al.* (2021) reported that the inclusion of 4%-SWA affected redness value of LL pork. The differences in the content of natural antioxidants such as polysaccharides and polyphenols present in the SWA, and some possible interactions between the polysaccharides present in the SWA, might affect the oxymyoglobin (Ortiz *et al.*, 2008) and therefore it influences meat color. Beef from pasture-raised animals tend to have higher levels of antioxidant compounds, like phenols, terpenoids, carotenoids and tocopherols (Van Vliet *et al.*, 2021) which also affect color stability.

The pH represents one of the most important characteristics of meat because it is highly related to meat quality. In this study, muscle pH values were not affected by treatments. Some authors reported that feeding has little influence on water holding capacity, which is an important parameter to define the taste and technological quality of meat (Braden *et al.*, 2007). In this study, the water holding capacity expressed as total cooking losses were not affected by the inclusion of SWA in the diet of finishing steers. Previous studies carried out with the same additive, but in pigs (Jerez-Timaure *et al.*, 2021), also reported that the 2 and 4% inclusion of SWA does not affect the water holding capacity.

Postmortem pH directly affects quality characteristics, such as water holding capacity, color, and, to a lesser degree, tenderness (Jacob & Hopkins, 2014). In addition, the speed of the decrease in pH in the carcass is influenced by multiple factors or *antemortem* handling of the animal (Gallo *et al.*, 2013; Gallo & Huertas, 2016) or *postmortem* factors (Jacob & Hopkins, 2014; Ponnampalam *et al.*, 2017). For decades, GPOT has been used as a fair approximation for the total compounds transformable to lactic acid present in the muscle at slaughter (Monin & Sellier, 1985). It has been shown that anaerobic glycolytic processes occur early after exsanguination and these drastic biochemical changes are caused by the fast glycolytic activity that is triggered by the *postmortem* action of glycolytic enzymes.

Muscular glycogen content was very low at 24 h *postmortem*. Steers used in this study were fed mainly with forage and were fasted for almost 12 h before slaughtering, which may explain the low levels of MGC. However, pH values were ranged in the normal pH threshold (<5.8). Values greater than 5.8 are associated with dark cutting beef (Ponnampalam *et al.*, 2017; Apaoblaza *et al.*, 2015). Previous studies performed in southern Chile also reported low levels of *postmortem* MGC. Amtmann *et al.* (2006) found that *longissimus* muscle in grass-fed carcasses with pH <5.8 had 35.5 ± 15.7 mmol/kg of MGC. Meanwhile, Apaoblaza *et al.* (2015) reported MGC at 24 h of 6.34 ± 0.05 in normal pH carcasses.

Our results show that muscle glycogen, lactate, and glucose content, evaluated at 24 h *postmortem*, were not affected by the effect of the inclusion of the seaweed additive in the diet ($P>0.05$). In addition, they indicate that the *postmortem* glycolytic metabolism was similar in both groups of carcasses. The glycolytic potential used to determine the ability to convert glycogen into lactate (Monin & Sellier, 1985) was similar in both groups ($P>0.05$), indicating that the seaweed inclusion treatment did not modify the *postmortem* glycolytic capacity, allowing a normal drop of muscle pH in early *postmortem*.

The lipid fraction and fatty acid composition are mainly influenced by three factors: the age of the animal, the composition of the animals' diet, and the breed. Mwangi *et al.* (2019) stated that notable modifications produced by the feeding systems in the chemical composition of the meat are mainly in the fat content. In this study, meat from steers that were fed with 2%-SWA decreased its amount of total lipids. Natural pigments of brown marine algae, like fucoxanthin is found, which could inhibit the differentiation of preadipocytes into adipocytes. This has also been demonstrated by other studies with other carotenoids that, like fucoxanthin, have an allenic group in their composition (Quitral *et al.*, 2012). Also, it is known that marine algae are rich in vitamin A (Quitral *et al.*, 2012), which have retinoic acid in their composition, which restricts hyperplasia and/or regulates the growth hormone gene, resulting in a decrease in the fat deposition (Mwangi *et al.*, 2019). Seaweeds also have high amounts of vitamin D, being its active form (1,25-dihydroxyvitamin D₃), the cause of inhibiting the differentiation of adipocytes (Mwangi *et al.*, 2019). This occurs when its plasma levels increase, which occurs in situations of low dietary calcium intake (Mwangi *et al.*, 2019).

Given the growing demand for leaner meats or meats with lower fat content by consumers concerned about diet-health aspects, the use of the additive based on seaweed in cattle feed could become an alternative approach to achieve differentiated products with added value aimed at consumers who prefer lower lipid content in their protein foods of animal origin.

Morrill *et al.* (2017b) show that the meat of cattle fed with marine algae residues slightly modifies the fatty

acid profile of the beef, so it is necessary to evaluate the composition of fatty acids in this study because it could result in meats with a higher concentration of polyunsaturated fatty acids, due to the contribution of these compounds offered by brown seaweed. On the other hand, Casas *et al.* (2005) mentioned that the presence of omega 3 fatty acids in seaweed is an aspect of interest, since it could be an alternative to produce meats with a higher content of polyunsaturated fatty acids, which are beneficial for human health.

CONCLUSIONS

In general, no harmful effects were found in this study by feeding grass-fed steers during fattening with brown sea algae extracts; however, a reduction of total lipids present in the LL samples of grass-fed steers supplemented with 2%-SWA was evidenced. Since seaweed represents a group of organisms with diverse types of bioactive compounds, further studies are needed to understand the biological effects of this SWA on adipogenesis or fatty acid oxidation, as an alternative to producing beef with low content of fat.

ETHICS STATEMENT

Animal handling for animal care and welfare were performed and revised according to the Institutional Committee for Animal care and use of the Universidad Austral de Chile. Resolution number 281/2021. (<https://vidca.uach.cl/comite-bioetica-investigacion-uso-animales/>)

AUTHOR CONTRIBUTIONS

N.J-T.: Conceptualization, data curation and analysis, original draft preparation, and edition.

R.P.: conceptualization, investigation, writing, and review.

F.H. & J.F.: data collection, experimental work, investigation, and writing,

J.M.: supervision of data collection, data curation, data analysis, and review.

M.Br.: funding acquisition, conceptualization, review, and edition.

M.B.: experimental work, laboratory analysis, and revision of the manuscript.

All authors searched and reviewed the literature, discussed the contents of the manuscript, and approved submission.

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CONFLICTS OF INTEREST

The authors declare no conflict of interest.

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