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Effects on performance, carcass and meat quality of replacing maize silage and concentrate by grass silage and corn-cob mix in the diet of growing bulls

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ABSTRACT

Grass silage is barely used in intensive beef production, but it is unclear if its lower energy supply compared to maize-silage feeding really impairs growth performance. Diets with 100, 300, 500 or 750 g grass silage/kg dry matter replacing maize silage and concentrate were tested with or without dried corn-cob mix (CCM). Performance, carcass and meat quality were studied in 30 Limousin-sired bulls. Feeding grass silage, CCM, and concentrate in a ratio of 500:300:200 allowed to maintain a similar animal performance, carcass and meat quality compared to a conventional maize silage/concentrate diet. Increasing the dietary grass silage proportion to 750 g/kg decreased the shear force of the meat. The proportion of *n*-3 fatty acids in intramuscular fat increased with dietary grass silage proportion. Consequently, a strategic combination of grass silage with energy-rich forages may facilitate grassland-based feeding strategies in intensive beef production with favourable meat fatty acid profiles and a performance comparable to that with maize-silage based diets.

1. Introduction

Intensive beef production in Europe is characterised by high average daily gains (ADG) and aims at well-conformed carcasses with an evenly distributed fat cover. In Switzerland, the relatively low target slaughter weight of 520 to 550 kg should be reached within a fattening period of approximately 15 months after birth (Morel et al., 2017). To meet these aims, mainly entire male dairy × beef breed crossbred calves are fattened using maize-silage based diets. These diets are complemented with considerable amounts of concentrate to increase the energy density of the diet and to balance diets according to the requirements for metabolisable protein.

In contrast, high proportions of grassland-based feeds are scarcely used as an impaired growth and slaughter performance is expected due to their limited energy density (Juniper et al., 2005; Keady, 2005). However, using grassland-based feeds complies better with the ruminants' ability to convert non-digestible biomass for humans into high-quality protein. In addition, this type of feed is considered as one of

the key strategies for more sustainable ruminant production systems (Schader et al., 2015). In Switzerland, where grassland accounts for 70% of the agricultural land (BLW, 2020a), farmers participating in the governmental programme for 'grassland-based milk and meat production' (GMF; BLW, 2020b) receive subsidies when feeding diets consisting of ≥750 g/kg of grassland-derived feeds (on dry matter (DM) basis) and ≤ 100 g/kg of concentrate.

However, higher dietary levels of grassland-based feeds such as grass silage can only be regarded as a competitive feeding strategy in intensive beef production if a sustained growth performance and desired carcass and meat quality can be ensured. In this context, feeding grass silage instead of maize silage could improve the protein supply provided by the basal diet, consequently lowering the demand for additional protein sources (Huuskonen, Huhtanen, & Joki-Tokola, 2014). Particularly the use of imported soybean meal, one of the main protein sources used for ruminants, is often controversially discussed. Still, the high rumen degradability of the grass silage protein may result in limited amounts of extra metabolisable protein, even though earlier findings have shown

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only small effects on fattening performance of protein supplementation when feeding grassland-based diets (Huukskonen et al., 2014; Huukskonen & Huhtanen, 2015; Keller et al., 2021). An added value of feeding grass-silage based diets consists in beef lipids richer in *n*-3 fatty acids (FA) and with a favourably lower *n*-6/*n*-3 FA ratio (O'Sullivan et al., 2002; Staerfl, Soliva, Leiber, & Kreuzer, 2011), which is desirable from a human nutrition point of view (Simopoulos, 2002). However, this may result in changes in sensory quality with occurrence of fishy or grassy off-odours, which could impair consumer acceptance (Scollan et al., 2006; Wood et al., 2003). Higher vitamin E contents of the meat from animals fed grass silage might limit this problem by preventing lipid oxidation (O'Sullivan et al., 2002; Warren et al., 2008) and further enhance the nutritional value of the meat. The antioxidant properties of vitamin E may also help to maintain meat colour during display at retail (O'Sullivan et al., 2002; Warren et al., 2008).

Nevertheless, the energy content is the dietary factor in beef production that mostly determines the fattening performance (Huukskonen & Huhtanen, 2015). To enhance the energy density of grass-silage based diets without extra concentrate, forages rich in energy are needed to complement the grass silage. Such a feed, being even more energy dense than maize silage, is corn-cob mix (CCM), *i.e.*, ensiled or dried maize kernels plus cobs, which is allowed for intensive beef production in the Swiss GMF programme.

Therefore, the aim of this study was to investigate the extent to which the proportion of grass silage can be increased in the diet of intensively fed beef cattle and to quantify how carcass and meat quality are concomitantly affected. The following hypotheses were tested: (1) Increasing the dietary proportion of grass silage reduces growth and slaughter performance. (2) This decline can be completely or partially avoided by replacing the remaining maize silage by a more energy-dense forage. (3) The physicochemical and sensory meat quality is negatively affected (older animals at slaughter with lower tenderness) by an increasing grass silage proportion, but (4) the meat lipids of animals fed higher dietary proportions of grass silage provide a higher proportion of *n*-3 FA.

As an energy-dense forage, dried CCM was chosen, and a low-protein concentrate replaced the common high-protein concentrate used in maize-silage dominated diets. With this, the anticipated limitation of the diet in energy at sufficient (crude) protein supply from the grass silage compared to the maize silage was considered.

2. Animals, materials, and methods

2.1. Diets and animals

The experiment was approved by the Cantonal Veterinary office of Zurich, Switzerland (license no. ZH129/18), and carried out at the AgroVet-Strickhof research station (Lindau, Switzerland).

Five different diets were tested in the experiment. The control diet (G100) was composed of minimal grass silage, and mainly of maize silage and concentrate (ratio of 100:600:300 on a DM basis). A commercial high-protein beef cattle concentrate was employed (M-2252 Beef 25%, Meliofeed, Herzogenbuchsee, Switzerland). The main components of this concentrate were soybean meal, maize kernel, distillers' grain, rape seed cake, wheat, maize gluten, and wheat bran. The control diet was calculated to be sufficient to support ADG of 1.40 kg according to the Swiss nutrient recommendation for beef cattle (Morel et al., 2017). The four other diets were characterised by gradually increasing grass silage proportions. Two diets had ratios of grass silage, maize silage and concentrate of 300:500:200 (G300) and 500:300:200 (G500), respectively. The two remaining diets consisted of grass silage, CCM and concentrate at ratios of 500:300:200 (G500_{CCM}) and 750:150:100 (G750_{CCM}), respectively, and did not contain any maize silage. Different from the control diet, the four experimental diets with higher grass silage proportions included a commercial low-protein concentrate (M-2256, Meliofeed) which was composed of wheat, wheat bran, maize

kernel, molasses, and fat of animal origin (cattle and pig). The proximate composition of and nutrient supply by the experimental feeds are shown in Table 1. The ingredient composition of the diets is listed in Table 2.

Thirty Limousin × dairy breed crossbred bulls (dam breeds: ten Brown Swiss, eight Swiss Fleckvieh, eight Red Holstein, and four Holstein) initially weighing 164 ± 18 kg (mean ± standard deviation) and averaging 4.4 ± 0.4 months of age were allocated to the five experimental diets. When allocating bulls to diets, initial body weight (BW), sire (*n* = 16) and dam breed were considered to achieve balanced means across diets. Bulls of the different dietary treatments were randomly distributed to three pens with ten animals each. Electronic gates and transponders (Waagen Doehrn, Wesel, Germany) were used to permit bulls access to only their own feeding trough. Each pen provided a lying area with straw that was added freshly three times per week, as well as a feeding and running area with access to an outside area. The individual concentrate allowance was adjusted every 2 weeks to the measured *ad libitum* silage intake to be able to maintain the defined silage-to-concentrate ratio throughout the whole experiment. Grass silage, harvested as second and third cut from ryegrass-dominated clover mixed swards at the beginning of ear emergence, and maize silage harvested at half milk-line stage were stored in two bunker silos each that were switched in experimental weeks 29 and 10, respectively. The CCM was from one batch, while two different batches of each concentrate type were used (both switched in experimental week 23). Grass and maize silage (diets G100, G300 and G500) as well as grass silage and CCM (diets G500_{CCM} and G750_{CCM}) were mixed every other day, exchanged daily and offered *ad libitum*. In the second half of the experiment, concentrates were offered three instead of two times per day to limit intake per meal as absolute amounts were increasing. The high-protein concentrate (diet G100) included, per kg, calcium, 17 g, phosphorus, 6 g, magnesium, 4 g, sodium, 3 g, iron, 40 mg, zinc, 135 mg, manganese, 65 mg, copper, 17 mg, iodine, 1.35 mg, selenium, 0.2 mg, cobalt, 0.6 mg, vitamin A, 20'000 IE, vitamin D₃, 2'000 IE, vitamin E, 50 mg. The other diets were supplemented directly with 75 g/day and animal of a mixture providing, per kg, calcium, 93 g, phosphorus, 44 g, magnesium, 57 g, sodium, 183 g, chloride, 158 g, sulphur, 3 g, zinc, 1.67 g, manganese, 1.33 g, copper, 400 mg, iodine, 73 mg, selenium, 15 mg, cobalt, 33 mg, vitamin A, 400'000 IE, vitamin D₃, 80'000 IE, vitamin E, 667 mg. The increase in calcium requirements with elevated grass silage proportion were balanced by adding calcium carbonate (385 g Ca/kg) in varying amounts to the diets with increased grass silage proportions following Morel et al. (2017). Free access to NaCl-licking blocks and fresh water was provided at any time.

Every other week, the BW was recorded using a cattle scale (Ixonix FX 15, Texas Trading, Windach, Germany). Individual feed intake was measured on two consecutive days by weighing the amounts of feed offered and leftovers after 24 h. Grass silages, maize silages, CCM and concentrates were sampled in total 14, 15, 12 and 4 times, respectively. Silage samples were dried at 60 °C for 48 h and all feeds were ground in a centrifugal mill (ZM 200, Retsch GmbH, Haan, Germany) to pass a 1-mm sieve.

2.2. Slaughter, carcass quality assessment and sampling

According to common practice for the Swiss beef label Terra Suisse (www.migros.ch/de/einkaufen/migros-marken-und-labels/terrasuisse.html), bulls were slaughtered at a BW of 520 kg. After being fasted overnight for about 12 h, bulls were transported within approximately 30 min to the abattoir of the University of Zurich (Zurich, Switzerland). Due to differences in fattening periods necessary to reach the target BW, bulls were slaughtered on six different days within 99 days. At slaughter, bulls were on average 13.9 ± 1.5 months old. They were stunned with a captive bolt gun, followed by exsanguination *via* throat cut. Carcasses, organs (heart, liver, kidneys, spleen) and perirenal fat were weighed within 30 min after stunning. Dressing percentage was calculated as the percentage of hot carcass weight to final body weight that was measured

Table 1

Proximate composition and selected fatty acids (FA) of grass and maize silage, corn-cob mix (CCM) and concentrates.

Feed	Grass silage		Maize silage		CCM	Concentrates	
	1–28 ¹	29–46 ¹	1–9 ¹	10–46 ¹	1–46	High-protein	Low-protein
<i>n</i>	9	5	4	11	12	4	4
Proximate contents (g/kg DM)							
DM (g/kg wet weight)	350	363	381	376	913	893	888
Organic matter	879	891	962	965	983	904	966
Crude protein	154	137	83.0	74.3	72.2	269	140
Ether extract	35.7	30.1	33.2	33.8	44.4	74.2	42.1
Neutral detergent fibre	536	492	436	420	497	303	407
Acid detergent fibre	401	345	302	279	136	116	115
Acid detergent lignin	72.7	46.8	45.4	41.2	40.8	n.a.	n.a.
APDE ²	52.4	66.0	62.5	61.5	80.0	157	107
APDN ²	97.0	86.6	51.8	46.0	47.1	196	98
FA (g/100 g total FA)							
<i>n</i>	3	2	2	3	5	4	4
C12:0	0.561	1.158	0.398	0.490	0.025	0.102	0.057
C14:0	1.247	0.520	0.429	0.282	0.042	0.160	0.820
C15:0	0.220	0.156	0.076	0.049	0.011	0.049	0.154
C16:0	18.9	15.9	13.6	13.8	11.8	11.4	17.2
C16:0 <i>iso</i>	0.816	1.665	0.209	0.186	0.027	0.035	0.116
C16:1 <i>n</i> -7	1.480	0.213	0.251	0.182	0.095	0.117	0.665
C17:0	0.201	0.177	0.162	0.124	0.073	0.115	0.310
C17:1	0.170	0.158	0.083	0.071	0.045	0.050	0.196
C18:0	1.74	1.38	2.54	2.20	1.83	23.54	7.22
C18:1 <i>cis</i> -9	2.3	2.6	29.1	25.6	30.4	17.3	24.1
C18:1 <i>cis</i> -11	0.508	0.465	0.669	0.669	0.610	1.256	1.056
C18:2 <i>n</i> -6	14.3	16.2	44.1	48.3	52.2	39.0	41.9
C18:3 <i>n</i> -3	53.48	55.45	5.35	5.71	1.67	5.27	3.62
C20:1 <i>n</i> -9	0.136	0.129	0.252	0.273	0.232	0.272	0.480
C20:5 <i>n</i> -3	0.103	0.091	0.071	0.052	0.039	0.037	0.026
C22:0	0.575	0.669	0.427	0.308	0.148	0.400	0.141
C24:0	0.753	0.835	0.695	0.474	0.207	0.162	0.151
Σ Saturated FA	26.0	23.7	19.0	18.2	14.5	36.2	26.5
Σ Monounsaturated FA	5.5	4.2	31.2	27.4	31.4	19.3	27.7
Σ Polyunsaturated FA	68.5	72.0	49.8	54.4	54.1	44.5	45.8
Σ <i>n</i> -3 FA	53.76	55.70	5.44	5.77	1.71	5.31	3.70
Σ <i>n</i> -6 FA	14.6	16.2	44.2	48.5	52.2	39.1	42.1

APDE: metabolisable protein derived from ruminal available energy; APDN: metabolisable protein derived from ruminal protein fermentation; DM: dry matter; n.a.: not analysed.

¹ Periods of experimental weeks fed.

² APDE and APDN of grass silage, maize silage and CCM are estimated values according to [Agroscope \(2017\)](#) and those of concentrates were taken from manufacturer's declaration.

the morning one day before slaughtering.

Carcass conformation (C = excellent, X = poor) and fatness scores (1 = too lean, 3 = optimal and homogenous fat cover, 5 = excessively fat) were classified according to the Swiss classification system CH-TAX ([Proviande, 2015](#)) by an independent professional. This system is equivalent to the EUROP classification system.

At 24 h *postmortem* (*p.m.*), samples of the *Musculus longissimus thoracis et lumborum* (LTL) were excised from the left carcass side between the 8th and 13th rib. From the caudal side, a 2 cm-thick slice was cut, and adhering adipose and connective tissue were removed before homogenisation in a common household blender (Moulinette type DP-700, Moulinex, Ecully, France). The homogenised samples were vacuum packed and stored at -20°C for later analysis of meat proximate composition and FA profile. The remaining sample was weighed, vacuumed and aged for 21 days at 4°C in the dark. Subcutaneous fat samples were collected from the back of the left carcass, blended, vacuum packed and stored at -20°C for later analysis of oxidative stability.

2.3. Proximate analysis of the feeds and meat composition

Chemical analysis of feed and homogenised LTL samples was carried out following standard methods ([AOAC International, 1997](#); [VDLUF, 2012](#)). A gravimetric device model (TGS 701, Leco Corporation, St. Josephs, MI, USA; AOAC Official Method 942.05) was used for assessing DM and total ash. Nitrogen (N) content was analysed with a C/N-analyser (TruMac CN, Leco; AOAC Official Method 968.06) and CP

was calculated as $6.25 \times \text{N}$. Ether extract was determined using a Soxhlet extractor (extraction system B-811, Buechi, Flawil, Switzerland; AOAC Official Method 963.15). Intramuscular fat was determined after HCl hydrolysis (Hydrolysis Unit B-425, Buechi) according to [Mueller et al. \(2020\)](#). In feed samples, ash-corrected neutral detergent fibre (NDF; with heat-stable α -amylase from Sigma-Aldrich, St. Louis, MO, USA) and acid detergent fibre (ADF) were determined with the Fibertherm FT 12 (Art. 13–0026, Gerhardt, Koenigswinter, Germany; VDLUFA methods 6.5.1 and 6.5.2, respectively). Following ADF-analysis, acid detergent lignin (ADL; VDLUFA method 6.5.3) was analysed in silages and CCM by incubation with sulphuric acid (720 mL/L) for 3 h. Values of metabolisable protein resulting from rumen-undegraded protein and microbial protein synthesised from ruminal available energy (APDE) as well as metabolisable protein resulting from rumen-undegraded protein and microbial protein synthesis from ruminal available CP (APDN; [Daccord, 2017](#)) were estimated based on proximate nutrient contents according to [Agroscope \(2017\)](#).

Vitamin E (tocopherols and tocotrienols) was quantified in fresh and 21-day aged meat as described by [Gebenstein and Frank \(2012\)](#). Briefly, ascorbic acid (1% in ethanol, wt/vol) and water were added to meat samples, which then were saponified with saturated potassium hydroxide at 70°C for 30 min. Samples were cooled on ice and butylated hydroxytoluene (25 μL of a 1 mg/mL ethanolic solution) and glacial acetic acid were added. After double extraction with *n*-hexane, supernatants were pooled, dried by evaporation (Christ SpeedDry; Christ), the residual resuspended in methanol/water (85:15, vol/vol), and injected

Table 2

Effect on feed intake, growth, and slaughter performance of replacing maize silage and concentrate by grass silage (G) and corn-cob mix (CCM) in the diet of growing bulls.

Diet	G100	G300	G500	G500 _{CCM}	G750 _{CCM}		
Grass silage (g/kg diet DM)	100	300	500	500	750		
Maize silage (g/kg diet DM)	600	500	300	0	0		
CCM (g/kg diet DM)	0	0	0	300	150		
High-protein concentrate (g/kg diet DM)	300						
Low-protein concentrate (g/kg diet DM)		200	200	200	100		
<i>n</i>	5	5	6	6	6	SEM	<i>P</i> -value
Days on experimental feed	247 ^a	314 ^b	300 ^b	270 ^a	305 ^b	11.1	***
Age (months)							
Age at start ¹	4.26	4.25	4.44	4.41	4.28	0.289	n.s.
Age at slaughter	12.5 ^a	14.7 ^b	14.4 ^b	13.4 ^a	14.5 ^b	0.27	***
Body weight (BW, kg)							
At start	170	163	160	166	161	12.4	n.s.
At slaughter	522	523	519	524	518	5.8	n.s.
Average BW gain (kg/day) ¹	1.43 ^b	1.15 ^a	1.20 ^a	1.34 ^b	1.17 ^a	0.055	***
DM intake (DMI; kg/day)							
Total	6.66	6.45	6.48	6.99	6.62	0.230	n.s.
Forage	4.71 ^a	5.21 ^{ab}	5.22 ^b	5.62 ^{bc}	5.96 ^c	0.291	***
Concentrate	1.95 ^c	1.24 ^b	1.26 ^b	1.37 ^b	0.67 ^a	0.062	***
Feed conversion ratio (kg DMI/kg BW gain)	4.66 ^a	5.62 ^b	5.38 ^b	5.23 ^b	5.65 ^b	0.207	***
Nutrient intake							
Crude protein (g/day)	926 ^c	702 ^a	797 ^b	861 ^{bc}	894 ^c	36.0	***
APDE (g/day) ¹	590 ^c	447 ^a	444 ^a	517 ^b	439 ^a	21.7	***
APDN (g/day)	632 ^d	451 ^a	512 ^b	559 ^{bc}	571 ^{cd}	23.4	***
Hot carcass weight (kg) ¹	294	287	288	296	289	5.5	n.s.
Dressing percentage	56.4	54.8	55.6	56.5	55.8	0.70	n.s.
Conformation score ^{2, 3}	4.0	3.9	4.0	4.2	3.6	0.32	n.s.
Fat cover score ^{2, 4}	2.0	2.6	2.8	2.5	2.7	0.32	n.s.
Organ weights (g/kg carcass weight)							
Heart	6.37	7.53	6.89	7.13	7.06	0.316	°
Liver	19.8	19.4	19.5	20.2	19.4	0.56	n.s.
Spleen	3.52	3.50	3.38	3.84	3.33	0.269	n.s.
Kidneys	3.58	3.42	3.55	3.58	3.84	0.234	n.s.
Perirenal fat (g/kg carcass weight)	14.1	19.0	18.3	13.2	15.5	3.38	n.s.

APDE: metabolisable protein resulting from ruminal available energy; APDN: metabolisable protein resulting from ruminal protein fermentation; DM: dry matter; n.s.: not significant; SEM: standard error of the mean.

Significance of difference is indicated as *** $P < 0.001$, ° $P < 0.10$ and n.s. = not significant.

^{a,b,c} Means carrying different superscripts within variable are different at $P < 0.05$.

¹ Data was transformed for statistical analysis but means of untransformed data are presented.

² Data was analysed using Kruskal Wallis test for nonparametric data.

³ Defined as 1 = poor and 5 = excellent according to CH-TAX classification.

⁴ Defined as 1 = too lean, 3 = optimum, evenly covered with fat, 5 = excessively fat according to CH-TAX classification.

into a Jasco HPLC system (AS-950 Plus autosampler, PU-980) equipped with a Kinetex PFP column (2.6 μm , 150 \times 4.6 mm; Phenomenex, Aschaffenburg, Germany). A methanol-water solution (85:15, vol/vol) was used as the mobile phase. Tocopherols and tocotrienols were detected at an excitation wavelength of 296 nm and emission wavelength of 325 nm. Recording and integration of peaks was done using ChromPass version 1.8.6.1 (Jasco). For quantification of tocopherols and tocotrienols, peaks were compared to external standard curves of authentic compounds.

2.4. Analysis of physicochemical meat quality and fat shelf life

Temperature and pH of the LTL were measured 24 h *p.m.* between the 8th and 9th rib using a combined pH-/thermometer (testo 205, Testo Ltd., Alton, Hampshire, United Kingdom). The device was calibrated using DuraCal pH buffer solutions (pH 4 and 7; Hamilton Company, Bonaduz, Switzerland) and the integrated temperature sensor ensured precise temperature compensation. Ageing loss was determined on day 21 *p.m.* after gently blotting dry and weighing the sample. The CIE L*a*b* colour space (lightness, redness, and yellowness, respectively) was measured with a Chroma Meter (model CR-400 with illuminant C, 2° standard observer, 8 mm aperture; Konica Minolta, Tokyo, Japan) on in total five points on two slices of 1 cm thickness each. The slices were cut from the caudal side of the LTL sample and let bloom (freshly cut side on top) for 60 min at 4 °C in the dark. Another four

slices of 2 cm each were dissected from the caudal side and weighed. To determine drip loss according to Honikel (1998), two slices were placed in a two-layer net each and hung in a sealed plastic bag for 24 h at 4 °C. The other two slices were vacuum packed and cooked in a water bath at 75 °C until a core temperature of 72 °C was reached, which was controlled with a digital thermometer (testo 108, Testo Ltd., Alton, Hampshire, United Kingdom). Always four to six samples were cooked at the same time in 11 cooking batches in total. After cooling samples in cold tap water, the slices were blotted dry and weighed to assess cooking loss. The cooked slices were then kept at room temperature and seven to ten cylindrical cores with a diameter of 1.27 cm were drilled along the muscle fibres using a cork borer. Shear force was measured perpendicular to the muscle fibres with a V-shaped Warner-Bratzler shear force blade mounted on a texture analyser (ProLine table-top machine Z005, Zwick Roell, Ulm, Germany) with a shear plate thickness of 3 mm, a load cell of 5 kg, and a crosshead speed of 400 mm/min. For all variables, values were averaged per animal for statistical analysis.

Oxidative stability in homogenised samples of subcutaneous fat was determined using Rancimat (model 697, Metrohm, Herisau, Switzerland) at 110 °C and an airflow of 20 L/h after melting the fat at 80 °C for 60 min and sieving it through a common kitchen sieve to remove connective tissue.

2.5. Fatty acid analysis in feeds and meat

For FA analysis in feed and homogenised meat samples, total lipids were extracted with hexane:isopropanol (HIP) in a ratio of 3:2 (vol/vol) using an accelerated solvent extractor (ASE 200, Dionex Corporation, Sunnyvale, CA, USA) for dietary lipids and by direct dispersion of meat lipids in HIP (Polytron® model PT 6000, Kinematica AG, Luzern, Switzerland). Methanolic NaOH and BF₃ (IUPAC, 1987) were added under cooking conditions to convert FA to FA methyl esters (FAME) as outlined by Wolf, Ulbrich, Kreuzer, Berard, and Giller (2018). As an internal standard, 5 mg C11:0 triglyceride (Fluka Chemie, Buchs, Switzerland) was added, followed by adding methylation reagents. Samples were analysed using a gas chromatograph (HP 6890, Agilent Technologies, Inc., Wilmington, PA, USA) equipped with a CP7421 column (wall-coated open tubular fused silica, 200 m × 0.25 mm × 0.25 µm; Varian, Lake Forest, CA, USA). At a split rate of 1:20, 1 µL of FAME was injected. Hydrogen was used as carrier gas at a flow rate of 1.7 mL/min. The detailed temperature conditions are described in Wolf et al. (2018). Detector temperature was 270 °C. The chromatograms were compared to a common 37-component standard (Sigma Aldrich, Steinheim, Germany) for identification of FA, whereas quantification of FAME peak areas was done using the HP ChemStation® software (Agilent, Palo Alto, CA, USA). Fatty acids are expressed as proportion of total FAME analysed.

2.6. Sensory analysis

For the sensory analysis, 2 cm thick deep frozen LTL meat slices were thawed for 24 h at 4 °C, blotted dry and stored at room temperature for around 1 h. Each slice was grilled at 170 °C for in total 5.5 min and flipped over after exactly 1.5, 3 and 4.25 min. Afterwards, slices were cut into pieces of approximately 2 × 2 cm. The cut samples were kept at 70 °C until sensory testing.

Seven members of a trained panel, consisting of Agroscope (Posieux, Switzerland) collaborators, participated in the sensory tests. Intensity of the attributes odour, tenderness, juiciness, fatty mouthfeel, umami, acidic, total flavour, rancidity, metallic, fishy and duration in mouth (aftertaste) was measured on an unstructured 10-cm line scale labelled from none (0) to high (10) intensity. Prior to data collection, panellists participated in two training sessions to get familiar with the attributes of interest. In each of the six test sessions, panellists evaluated two sample sets, each consisting of meat cuts of three different animals. Samples of each test set were randomised following a William Latin Square design. All samples were coded with three-digit random numbers. The sensory data was collected using the software FIZZ (version 2.51 Biosystèmes, Couternon, France).

White bread, still water and warm black tea were provided for neutralisation of the mouth between samples. Tests were conducted at room temperature under day light conditions in the sensory laboratory at Agroscope Posieux.

2.7. Statistical analysis

Two animals had to be excluded from the study. One bull (G100) had to be slaughtered due to an abomasal displacement on experimental day 205. Another animal (G300) was excluded due to a very low performance (BW >60 kg below the target BW of 520 kg when all other experimental animals had been slaughtered). Therefore, the final group sizes for G100 and G300 were $n = 5$ instead of $n = 6$.

For data analysis, R version 4.1.2 (R Core Team, 2021) was used. Animal was considered as the experimental unit. Performance, slaughter, and physicochemical meat quality data were evaluated by analysis of variance for diet effects with the aov function. Initial BW was included in the model as covariate when analysing days on experimental diets, final BW, intakes of total feed, silage, concentrate and nutrients as well as feed conversion ratio. Initial BW was not considered in any other

performance-related variable as it was found to have no significant effect. Data of the sensory meat evaluation was subjected to a mixed model analysis using the lmer function. Diet and panellist were included as fixed effect, while tasting session and animal were considered as random effects. For multiple comparisons of means, a Bonferroni-applied *post hoc* test was used via the cld function. In case of data heteroscedasticity or non-normal distribution of residuals, data were transformed for statistical analysis as indicated in the tables. For data of conformation and fat cover score, pH_{24h postmortem}, cooking loss and C17:1, a Kruskal Wallis test was carried out for evaluating the diet effect, while a Bonferroni-adjusted *post hoc* analysis was applied by using the dunnTest function for multiple comparisons among means in case of non-parametric data. The lmer function was also used to evaluate vitamin E related data using diet and ageing time as fixed and animal as random factors. *Post hoc* analysis was done using the glht function to calculate contrasts within diets among ageing time or across diets within ageing time. Effects at $P < 0.05$ were considered statistically significant and effects at $0.05 < P < 0.1$ as trends. Data is presented as untransformed arithmetic means ± standard error of the mean (SEM).

3. Results

3.1. Feed and diet composition as well as growth and slaughter performance

Based on the proximate composition of the different dietary ingredients (Table 1) and their varying dietary proportions, total diets varied in their nutrient supply. The average CP contents calculated from the analysed data of the individual ingredients for diets G100, G300, G500, G500_{CCM} and G750_{CCM} were 140 ± 2.1 , 109 ± 3.4 , 123 ± 3.7 , 123 ± 3.0 , and 135 ± 4.7 g/kg DM, respectively. The corresponding contents of APDE were 89.1 ± 0.36 , 69.6 ± 1.23 , 68.8 ± 2.16 , 74.2 ± 2.61 , and 65.1 ± 2.87 g/kg DM and those for APDN were 95.7 ± 1.46 , 70.1 ± 1.99 , 79.2 ± 2.15 , 80.1 ± 1.81 , and 86.1 ± 3.06 g/kg DM. The CP content of the low-protein concentrate was only about half of that of the high-protein concentrate. The FA profile clearly differed between the grass silage and the maize-based forages (Table 1), with C18:3 *n-3* accounting for more than half of the total FA found in grass silage. Further prevalent FA in grass silage were C16:0 and C18:2 *n-6 cis*, whereas the main FA found in the maize-based forages were C18:2 *n-6 cis*, C18:1 *cis-9* and C16:0 in a descending order. In both concentrates, C18:2 *n-6 cis* was the predominant FA. The further major FA (C18:0, C18:1 *cis-9*, C16:0) varied in their proportions between the concentrates.

All groups had a similar average initial and final BW (521 ± 2.0), where the final BW closely coincided with the target BW at slaughter (Table 2). Starting into the experiment at the same age, animals of G100 and G500_{CCM} were younger ($P < 0.001$) at slaughter than the animals of all other groups as they needed less ($P < 0.001$) time (days on experimental diet) to reach the slaughter weight. Accordingly, G100 and G500_{CCM} bulls had a higher ($P < 0.001$) ADG than the bulls on the remaining diets. The diet effect on the BW became significant from week 15 onwards (Fig. 1). Average total DMI (6.60 ± 0.088 kg DM/day and bull) was comparable for all diets tested (Table 2). The differences in forage and concentrate intakes (both $P < 0.001$) among groups resulted only from the different amounts supplied when following the experimental protocol. The feed conversion ratio was more favourable ($P < 0.001$) for G100 than for all other diets. The CP intake was highest in G100 and G750_{CCM} and lowest for G300 ($P < 0.001$). Bulls from groups G300, G500 and G750_{CCM} ingested less ($P < 0.001$) APDE than those fed G500_{CCM} which also had a lower ($P = 0.044$) intake than those fed G100. The APDN intake also differed ($P < 0.001$) among diets and was highest in G100 as well as in G750_{CCM} (not different from G500_{CCM}) and lowest in G300. Carcass weights, dressing percentages, and conformation scores were similar in all groups. Although fat cover scores were not statistically different, the bulls of the experimental groups achieved numerically on average the ideal fat cover score of 3, whereas those of

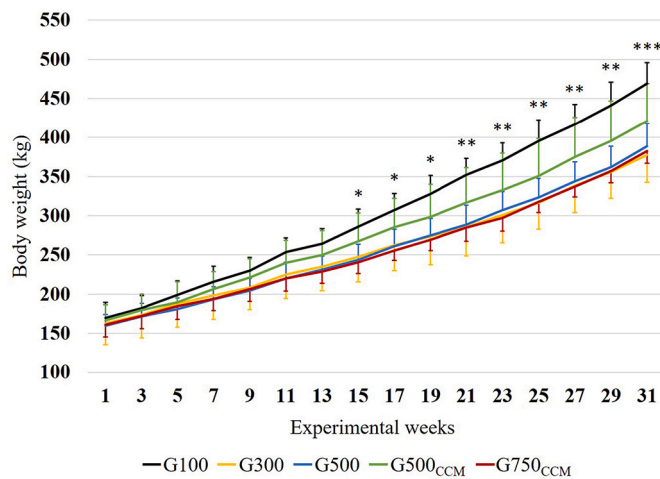


Fig. 1. Effect of replacing maize silage and concentrate by grass silage (G) and corn-cob mix (CCM) in the diet of growing bulls on their body weight development (arithmetic means \pm standard deviation). Diet type effect within experimental week, * $P < 0.05$; ** $P < 0.01$; *** $P < 0.001$.

the control group were mostly graded with 2. The dietary treatments did not significantly affect the proportionate weights of organs and perirenal fat.

3.2. Physicochemical quality of the meat

Carcass temperature and pH at 24 h *p.m.* did not differ among diets (Table 3). Water-holding capacity, determined as ageing, drip and

Table 3

Effect on physicochemical and sensory quality of the *longissimus thoracis et lumborum* aged for 21 days of replacing maize silage and concentrate by grass silage (G) and corn-cob mix (CCM) in the diet of growing bulls.

Diet (D)	G100	G300	G500	G500 _{CCM}	G750 _{CCM}	SEM	P-value
n	5	5	6	6	6		
pH _{24 h p.m.} ¹	5.97	5.75	5.89	5.75	5.98	0.167	n.s.
Temperature _{24 h p.m.} (°C)	2.88	3.82	3.15	3.25	3.59	0.419	n.s.
Water holding capacity (%)							
Ageing loss	1.75	2.33	2.29	2.11 ²	1.49	0.473	n.s.
Drip loss ³	1.05	1.35	1.30	1.23	1.07	0.232	n.s.
Cooking loss ¹	22.7	22.2	24.2	22.9	17.4	2.72	n.s.
Colour							
L* (lightness)	43.5	43.9	45.0	43.8	41.1	1.80	n.s.
a* (redness)	18.9 ^a	22.4 ^{ab}	21.7 ^{ab}	22.9 ^b	19.4 ^{ab}	1.19	*
b* ³ (yellowness)	12.2	13.8	13.7	14.4	11.5	1.05	*
Shear force (N) ³	75.2 ^b	51.6 ^{ab}	50.4 ^{ab}	56.2 ^{ab}	40.3 ^a	9.13	*
Chemical composition (g/kg)							
Moisture	752	751	753	751	746	5.5	n.s.
Protein	214	204	206	212	208	5.0	n.s.
Fat (ether extract) ³	7.6	13.5	11.7	7.5	7.5	2.44	°
Ash	16.8	14.3	15.1	17.2	14.7	1.49	n.s.
Oxidative stability (h)	1.82	1.93	2.46	1.91	1.55	0.246	°
Sensory attributes (0 (none) to 10 (high intensity))							
Odour intensity	5.63	5.26	5.08	5.51	5.14	0.325	n.s.
Tenderness	4.39	4.13	4.98	5.88	5.50	0.409	n.s.
Juiciness	4.33	4.74	5.20	5.29	4.74	0.353	n.s.
Fatty mouthfeel ³	1.63	2.08	2.01	1.94	1.81	0.324	n.s.
Umami	3.07	3.25	3.00	3.31	2.74	0.346	n.s.
Acidic ³	1.50	1.46	1.15	1.17	1.26	0.260	n.s.
Flavour intensity ³	4.85	5.25	4.79	5.20	4.51	0.308	n.s.
Rancidity ³	0.86	0.88	1.02	1.08	0.74	0.314	n.s.
Metallic ³	1.21	0.75	0.82	0.97	0.98	0.215	n.s.
Fishy ³	0.47	0.29	0.48	0.37	0.55	0.166	n.s.
Duration in mouth	5.05	5.07	4.80	4.87	4.53	0.262	n.s.

SEM: standard error of the mean. Significance of difference is indicated as * $P < 0.05$, ° $P < 0.10$ and n.s. = not significant.

^{a,b} Means carrying different superscripts within variable are different at $P < 0.05$.

¹ Data was analysed using Kruskal Wallis test for non-parametric data.

² n = 5.

³ Data was transformed for statistical analysis but means of untransformed data are presented.

cooking loss, remained unaffected by the diet. The meat of G500_{CCM} was redder ($P = 0.041$) than that of G100. Meat yellowness differed ($P = 0.035$) among diets but no distinct differences between means were revealed, and lightness remained unaffected. The shear force of the meat was lower for G750_{CCM} than for G100 ($P = 0.011$). Contents of moisture, protein and ash of the meat were unaffected by the diet, while intramuscular fat content tended to differ between groups ($P = 0.074$; numerically highest with G300 and G500). There was a trend ($P = 0.055$) for differences in oxidative stability of the subcutaneous fat in bulls, with the numerically highest level found with G500 and the lowest with G750_{CCM}. For α -tocotrienol, no diet and ageing effects were found (Table 4). A diet effect ($P = 0.009$) was found for γ -tocotrienol content which was highest in aged G100 meat. The α -tocopherol content tended to differ ($P = 0.099$) among diets and ageing led to a higher ($P = 0.017$) α -tocopherol content in G300 meat compared to fresh meat. The diet also affected ($P < 0.001$) γ -tocopherol content, which was highest in fresh G100 meat. After 21 days of ageing, meat γ -tocopherol content was higher in G100 than in G500, G500_{CCM}, and G750_{CCM} as well as in G300 and G500_{CCM} than in G750_{CCM}. Total vitamin E content remained unaffected by diet, while higher contents were found in G300 after ageing compared to fresh meat.

3.3. Sensory evaluation of the beef

The diet had no impact on the sensory perception of the beef steaks (Table 3). However, animals fed G500_{CCM} were graded with numerically higher mean values for the two texture attributes tenderness and juiciness compared to animals fed G100, G300 and G500, G750_{CCM}, whereas no dietary impact was observed for the flavour attributes. Sensory attributes were evaluated differently (for all $P < 0.001$) among panellists

Table 4

Tocotrienol, tocopherol and total vitamin E content of fresh or 21-day aged *longissimus thoracis et lumborum* when replacing maize silage and concentrate by grass silage (G) and corn-cob mix (CCM) in the diet of growing bulls.

Diet (D)	G100		G300		G500		G500 _{CCM}		G750 _{CCM}		SEM	P-value		
	0	21	0	21	0	21	0	21	0	21		D	A	D × A
Ageing period in days (A)	0	21	0	21	0	21	0	21	0	21				
n	5		5		6		6		6					
Tocotrienols (µg/kg)														
α-tocotrienol	40.0	42.1	40.2	35.2	35.1	37.8	44.6	37.7	35.8	38.5	9.39	n.s.	n.s.	n.s.
γ-tocotrienol	6.76	11.02 ^Y	5.38	4.48 ^X	4.79	4.73 ^X	5.67	4.67 ^X	5.02	4.42 ^X	3.286	**	n.s.	n.s.
Tocopherols														
α-tocopherol (g/kg)	1.51	1.67	1.78 ^a	2.28 ^b	2.05	2.18	2.21	2.43	2.12	2.06	0.276	°	*	n.s.
γ-tocopherol (µg/kg)	77.2 ^Y	73.5 ^{1z}	46.3 ^X	54.4 ^{yz}	29.3 ^X	33.2 ^{xy}	45.1 ^X	50.6 ^Y	22.8 ^X	21.0 ^X	8.66	***	n.s.	n.s.
Total vitamin E (g/kg)	1.64	1.83	1.87 ^a	2.38 ^b	2.12	2.26	2.31	2.54	2.19	2.12	0.283	n.s.	*	n.s.

Significance of difference is indicated as * $P < 0.05$, ** $P < 0.01$, *** $P < 0.001$, ° $P < 0.10$ and n.s. = not significant.

^{a,b} Means carrying different superscripts within variable and feeding group are different at $P < 0.05$.

^{x,y} Means carrying different superscripts within variable and fresh meat are different at $P < 0.05$.

^{xy} Means carrying different superscripts within variable and 21-day aged meat are different at $P < 0.05$.

¹ n = 4.

Table 5

Effect on fatty acid (FA) profile (g/100 g total FA) of the *longissimus thoracis et lumborum* of replacing maize silage and concentrate by grass silage (G) and corn-cob mix (CCM) in the diet of growing bulls.

FA ¹	G100	G300	G500	G500 _{CCM}	G750 _{CCM}	SEM	P-value
n	5	5	6	6	6		
Σ Fatty acids (mg/100 g tissue)	1006	1331	1529	1056	935	209.3	°
C14:0 ²	1.83	1.99	2.41	1.89	1.82	0.219	*
C14:1	0.484	0.564	0.584	0.357	0.407	0.0740	*
C15:0	0.260 ^a	0.352 ^{ab}	0.377 ^b	0.290 ^{ab}	0.377 ^b	0.0251	**
C15:0 iso	0.131 ^a	0.210 ^b	0.198 ^{ab}	0.143 ^{ab}	0.127 ^a	0.0224	**
C16:0 ²	22.8 ^a	24.0 ^{ab}	25.5 ^b	22.7 ^{ab}	22.5 ^a	1.05	*
C16:0 iso	0.200	0.239	0.210	0.197	0.208	0.0130	°
C16:1	2.68	2.98	3.21	2.29	2.43	0.272	*
C16:1 ×	0.327	0.378	0.397	0.299	0.412	0.0515	n.s.
C17:0 ²	0.601 ^a	0.728 ^{bc}	0.779 ^c	0.641 ^{ab}	0.893 ^c	0.1277	***
C17:0 iso	0.050 ^a	0.137 ^{ab}	0.208 ^{bc}	0.224 ^{bc}	0.274 ^c	0.0347	***
C17:1 ³	0.562 ^{ab}	0.718 ^c	0.694 ^{bc}	0.522 ^{ab}	0.649 ^{abc}	0.0244	***
C17:0 aiso	0.106	0.108	0.112	0.130	0.196	0.0325	°
C18:0	14.5	14.3	14.5	15.3	15.1	0.74	n.s.
C18:1 trans-6-8	0.181 ^b	0.174 ^b	0.155 ^{ab}	0.178 ^b	0.112 ^a	0.0207	**
C18:1 trans-9	0.275	0.260	0.245	0.251	0.238	0.0181	n.s.
C18:1 trans-10	0.400 ^b	0.312 ^{ab}	0.279 ^a	0.312 ^{ab}	0.241 ^a	0.0279	***
C18:1 trans-11	1.34	1.42	1.24	1.39	1.21	0.185	n.s.
C18:1 trans-12	0.290	0.287	0.275	0.308	0.252	0.0226	n.s.
C18:1 cis-9	32.4	33.1	32.7	28.4	29.2	1.80	°
C18:1 cis-10	0.184	0.205	0.221	0.241	0.232	0.0165	°
C18:1 cis-11	1.49	1.46	1.35	1.40	1.40	0.0742	n.s.
C18:1 cis-12	0.363	0.265	0.301	0.275	0.292	0.0683	n.s.
C18:1 cis-13	0.240	0.249	0.226	0.185	0.226	0.0295	n.s.
C18:2 trans-11, cis-15	0.065 ^a	0.104 ^{ab}	0.131 ^b	0.127 ^b	0.181 ^c	0.0151	***
C18:2 cis-9, trans-11 ²	0.541	0.450	0.389	0.381	0.356	0.0592	°
C18:2 n-6 ²	10.22 ^{ab}	7.77 ^{ab}	6.30 ^a	11.89 ^b	9.15 ^{ab}	2.081	*
C18:3 n-3 ²	0.67 ^a	1.10 ^{ab}	1.27 ^{ab}	1.97 ^{bc}	2.61 ^c	0.448	***
C20:0 ²	0.151	0.098	0.217	0.095	0.274	0.1641	n.s.
C20:3 n-6	0.637	0.477	0.370	0.569	0.628	0.0986	°
C20:4 n-6	3.01	2.44	1.97	3.16	2.97	0.542	n.s.
C20:4 n-3 ²	0.050 ^a	0.072 ^{ab}	0.088 ^{abc}	0.122 ^{bc}	0.169 ^c	0.0245	***
C20:5 n-3 ²	0.422 ^a	0.507 ^a	0.563 ^a	0.780 ^{ab}	1.257 ^b	0.1900	***
C22:4 n-6	0.408 ^b	0.260 ^{ab}	0.196 ^a	0.297 ^{ab}	0.224 ^a	0.0482	**
C22:5 n-3	0.87 ^a	0.87 ^a	0.95 ^a	1.20 ^{ab}	1.78 ^b	0.02287	**
C22:6 n-3 ²	0.110 ^a	0.127 ^a	0.145 ^a	0.149 ^{ab}	0.310 ^b	0.0635	**
Σ Saturated FA	41.1	42.7	45.0	42.1	42.3	1.41	°
Σ Monounsaturated FA	41.6	42.9	42.3	36.9	37.7	2.14	*
Σ Polyunsaturated FA ²	17.3	14.4	12.6	21.0	20.0	3.29	*
Σ n-3 FA ²	2.16 ^a	2.70 ^{ab}	3.04 ^{ab}	4.26 ^{bc}	6.16 ^c	0.891	***
Σ n-6 FA ²	14.5	11.1	9.0	16.2	13.2	2.78	*
n-6/n-3 FA ²	6.75 ^d	4.11 ^c	2.96 ^b	3.80 ^{bc}	2.14 ^a	0.434	***

SEM: standard error of the means.

Significance of difference is indicated as * $P < 0.05$, ** $P < 0.01$, *** $P < 0.001$, ° $P < 0.10$ and n.s. = not significant.

^{a,b,c} Means carrying different superscripts within variable are different at $P < 0.05$.

¹ Only fatty acids making up >0.15 g/100 g total FA are displayed; all others were considered as traces.

² Data was transformed for statistical analysis. Means of untransformed data are presented in the table.

³ Data was analysed using the nonparametric Kruskal Wallis test for multiple comparison of means and Bonferroni-adjusted Dunn test for *post hoc* analysis.

(data not reported).

3.4. Fatty acid profile of intramuscular fat

The diet tended to influence total FA content in LTL, which was numerically highest for G500 and lowest for G750_{CCM} (Table 5). The four major FA found in the meat lipids of all groups were C18:1 *cis*-9, C16:0, C18:0 and C18:2 *n*-6 *cis*, and this in descending order. The proportion of C18:1 *cis*-9 tended ($P = 0.057$) to differ among groups. The proportion of C16:0 was lower in G100 ($P = 0.039$) and G750_{CCM} ($P = 0.021$) compared to G500. No effect was found for C18:0 proportion. The proportion of C18:2 *n*-6 *cis* was lower ($P = 0.038$) for G500 than for G500_{CCM}. The highest ($P < 0.001$) proportion of C18:2 *trans*-11, *cis*-15 was found in meat lipids of G750_{CCM}, which was also higher in G500 ($P = 0.025$) and G500_{CCM} ($P = 0.005$) than in G100. A clear increase in the proportion with elevating dietary levels of grass silage was also found for C18:3 *n*-3 and C20:4 *n*-3 with the significantly lowest proportion for G100, and highest proportions in G500_{CCM} and G750_{CCM}. The proportions of C20:5 *n*-3, C22:5 *n*-3 and C22:6 *n*-3 were also significantly higher in G750_{CCM} than in the intramuscular fat of G100, G300 and G500. In contrast, G750_{CCM} resulted in a significantly lower proportion of C22:4 *n*-6 than the control group, this was also the case for G500. Additionally, proportions of C18:2 *cis*-9, *trans*-11 tended ($P = 0.052$) to vary and were numerically lowest in G750_{CCM}. There was a trend ($P = 0.10$) for sum of saturated FA (SFA) to be affected by the diet which was numerically highest for G500. Sums of mono- (MUFA) and polyunsaturated FA (PUFA) as well as the sum of *n*-6 FA differed significantly among groups. However, no significant differences between individual treatments were identified by the *post hoc* analysis for these FA. The sum of *n*-3 FA was higher in G500_{CCM} than in G100 ($P = 0.024$) as well as higher in G750_{CCM} than in G100 ($P < 0.001$), G300 ($P = 0.003$), G500 ($P = 0.009$). Overall, the total meat *n*-3 FA increased with the dietary proportion of grass silage. Consequently, the ratio of *n*-6/*n*-3 FA decreased in general with higher proportions of grass silage. The greatest ($P < 0.001$) *n*-6/*n*-3 FA ratio overall was found for G100, while the smallest overall ratio was found for G750_{CCM}.

4. Discussion

4.1. Diet composition and animal performance

The grass silage used in the present experiment was particularly rich in NDF and ADF compared to tabulated values of 356 g NDF/kg DM and 262 g ADF/kg DM given for silage material harvested from similar botanical composition and growth stage, while CP content was comparable (Feedbase, 2021). Lower fibre contents of the grass silage could have improved the competitiveness of the grass-silage based diets compared to the control diet in the present study. The proximate composition of the maize silage complied mostly to tabulated values, with a slightly higher ADF content compared to that specified in the feed tables (243 g/kg DM; Feedbase, 2021). In CCM, fibre contents were higher compared to reported values for dry ear maize without husks (Heuzé, Tran, & Lebas, 2019) or ensiled CCM (147 g NDF/kg DM, 61.4 g ADF/kg DM; Feedbase, 2021).

The observed differences in animal performance may be explained by the varying proportions of metabolisable energy (ME) and protein supply by the different experimental diets. The ME contents of the total diets (MJ/kg DM; measured by individual daily collection of total faeces and urine output on 7 days followed by 2 days of measurement of methane production) were 11.5 for G100, 11.3 for G300, 11.0 for G500, 11.5 for G500_{CCM} and 10.8 for G750_{CCM} (unpublished data). These data show that including higher proportions of grass silage in the diet reduced ME supply which could partly explain the lower performance. The substitution of maize silage by CCM (G500 vs. G500_{CCM}) counterbalanced the reduced ME supply and resulted in a comparable ME supply by diets G100 and G500_{CCM}. In terms of ME supply and growth

performance, diet G500_{CCM} was not only superior to G500, but even to G300. The impaired performance found with G500 and G750_{CCM} may therefore have resulted from the lower ME supply. However, using CCM in the diet with the highest grass silage level (G750_{CCM}) caused ME supply to approach that of G500, despite the higher grass silage and the lower concentrate proportion. This therefore prevented even higher losses in performance. Consequently, using CCM can partly or even fully compensate for the decline in ME supply when replacing maize silage and part of the concentrate by grass silage. On farm, ensiled CCM is more common than dried CCM. Ensiled material has a higher ruminal starch degradability (700 g/kg DM vs. 500 g/kg DM for dried CCM; Feedbase, 2021) which goes along with a reduced efficiency of ME utilisation due to higher energy losses by fermentation (methane and heat) compared to enzymatic starch degradation in the small intestine (Huhtanen & Sveinbjörnsson, 2006). This might limit the competitiveness of ensiled CCM compared to maize silage.

According to the Swiss feeding recommendations for ruminants (Daccord, 2017; Morel et al., 2017), APDE and APDN should be balanced to ensure an optimal metabolisable protein supply. This was achieved for G300 and mostly for G100 and G500_{CCM}, whereas APDN was higher than APDE in G500 and G750_{CCM}, indicating an excess of rumen-degradable protein due to high CP contents and a lack of energy available for ruminal microbial protein synthesis to utilise the entire CP. The decrease in performance with G300 compared to G100 was likely caused by a combination of a limited supply of energy and metabolisable protein. It remains unclear whether the supplementation with additional metabolisable protein would have compensated for the deficiency of this diet. The APDE intake was lower in G500_{CCM} than in G100 but still considered adequate to maintain a comparable performance level as confirmed by the present results.

Compared to Swiss standards for intensive beef production (Morel et al., 2017), only G100 and G500_{CCM} bulls achieved a satisfactory ADG of >1.3 kg. Reduced growth when feeding higher amounts of grass silage have been reported earlier (Juniper et al., 2005; Keady, Lively, Kilpatrick, & Moss, 2007). In contrast, feeding exclusively grass silage as forage resulted in a performance of growing bulls equivalent to that of bulls fed only maize silage as a forage in the study of Staerfl et al. (2011). However, concentrate type and allowance remained unchanged in the experiment conducted by Staerfl et al. (2011), whereas in the present experiment both, type and dietary proportion differed between some feeding groups. Although most performance-related variables were comparable for G100 and G500_{CCM}, any elevation of grass silage in the diet reduced the feed conversion ratio compared to the control, also in G500_{CCM}, indicating a slightly better performance of the control diet.

The comparable slaughter performance (carcass weight and dressing percentage) as well as the similar carcass quality (conformation and fat cover) likely resulted from the comparable final BW. Similar findings were also observed in other studies when varying dietary proportions of grass or maize silage (Juniper et al., 2005; Staerfl et al., 2011), also when combined with different concentrate proportions (French et al., 2000; Keady et al., 2007; Keady, Gordon, & Moss, 2013). Muscle formation is mostly genetically determined and will only decline in case of metabolic protein deficiency. Numerically, the G100 bulls as the only group did not meet the optimum score of 3 (evenly distributed fat cover). In practice, a score of 2 leads to a substantial reduction of the sales revenue. Overall, this finding coincides with the results of a recent farm survey conducted in Switzerland where only about two thirds of the carcasses produced under the same label (Terra Suisse) as the bulls in the present study met the score of 3 (Janett, Wyss, Favre, Scheurer, & Reidy, 2021).

4.2. Meat quality

The water-holding capacity of the meat was not affected by any of the diets which is consistent with findings by Keady, Gordon, and Moss (2013) for cooking loss and French, O'Riordan, et al. (2000) for drip loss. No effect, or no systematic effect with respect to dietary grass silage

proportion, on meat lightness, redness and yellowness was found in our study. The findings are partly in accordance with earlier studies in which increasing amounts of grass silage or grazed grass did not affect meat colour (French, O'Riordan, et al., 2000; Juniper et al., 2005; Keady et al., 2007). In contrast, Nuernberg et al. (2005) reported beef from grass-based feeding systems to be darker, while Warren et al. (2008) observed lighter and less red meat in 19-months old bulls fed grass silage compared to a concentrate-based diet. Besides, Warren et al. (2008) found meat yellowness to be slightly increased in 14-months old bulls fed grass silage instead of concentrate. A bright, cherry-red coloured meat is preferred by consumers and a^* -values ≥ 14.5 are perceived acceptable and associated with fresh beef of high nutritional quality (Holman, van de Ven, Mao, Coombs, & Hopkins, 2017; Holman & Hopkins, 2021). For lightness and yellowness, L^* - and b^* -values should be greater than, 31.4 and 6.3, respectively (Holman & Hopkins, 2021; Realini, Staincliffe, Taukiri, Craigie, & Farouk, 2018; Zhang et al., 2021). These thresholds were achieved among all dietary treatments. This indicates that the small but significant colour differences of meat from the different experimental groups would likely not affect consumer acceptance. However, the use of different illuminants (C in the present study vs. D_{65} in other studies) may slightly limit comparability between study results, though both illuminants simulate daylight. Different from meat colour, feeding higher proportions of grass silage to beef cattle may lead to yellower carcass fat (Keady et al., 2013; Moloney & Drennan, 2013) which limits the acceptance and suitability for the market where white fat is requested (Moloney & Drennan, 2013).

We found that shear force in the G750_{CCM} meat was almost only about half of that found in the G100 bulls which showed particularly high values. This is in line with an earlier study reporting a higher tenderness in beef with a lower $n-6/n-3$ FA ratio (Listrat et al., 2020). This relationship was however not observed in lambs (Ponnampalam et al., 2001; Ponnampalam, Sinclair, Egan, Blakeley, & Leury, 2001). To the best of the authors' knowledge, underlying pathways for the potential effect of the $n-6/n-3$ FA ratio on meat tenderness have not yet been published. Threshold values for acceptable tenderness measured as shear force vary between different studies (Holman & Hopkins, 2021). A range of 41–49 N seems to represent the threshold value until which consumers perceive beef as tender. Consequently, the meat of the G750_{CCM} group likely provided an acceptable eating quality, whereas the meat of all other groups must be considered as tough (>50 N). Tenderness as perceived by the sensory panel, though not significantly different, pointed into the same direction in a numerical sense as found with the shear force measurements. Some other studies did not report any effects of grass silage proportion and concentrate level on meat shear force when the meat was aged for at least 7 days (French, O'Riordan, et al., 2000; Keady et al., 2013), whereas Nuernberg et al. (2005) found higher shear force values in animals fed grass-based feeds compared to those fattened on concentrate. There are three major factors influencing shear force: Genetics, age at slaughter (Purchas, Burnham, & Morris, 2002), which represents the only factor determined by nutrition, and finally carcass processing conditions. The first and the last factor were kept constant as far as possible, but age at slaughter was higher with G750_{CCM} than with G100, a phenomenon also used as an explanation by Nuernberg et al. (2005). However, in the present study, the relationship was opposite regarding shear force measurements, which can be explained rather by involuntary genotypic differences in the animals allocated to the respective groups than an effect of age. There were only unsystematic differences among groups in intramuscular fat content even though there was an overall diet type effect. Likewise, French, O'Riordan, et al. (2000) found no effect on proximate muscle composition when feeding various forage types and concentrate levels to beef cattle, whereas in meat of grass-fed bulls intramuscular fat content was lower compared to concentrate feeding (Nuernberg et al., 2005). Like carcass fat cover, intramuscular fat content responds to energy supply. However, both indicators had no clear relationship to grass silage proportion and the associated decline in dietary energy

content.

A diet effect was found for the γ -tocotrienol and γ -tocopherol contents of the meat which decreased with an increasing proportion of grass silage in the diet. In contrast to previous reports, no diet effect was found for α -tocopherol and total vitamin E with increasing proportions of grass silage in the diet (O'Sullivan et al., 2002; Warren et al., 2008). Higher vitamin E contents would improve the nutritional value of beef for human consumption and potentially enhance oxidative stability of meat lipids, which was not investigated in the present study. However, for the oxidative stability of the subcutaneous fat, we observed a trend. A higher number of animals per treatment might have resulted in a more distinct effect. In the present study, the lowest absolute oxidative stability was found for G750_{CCM} and highest for G500. This complies not only with the vitamin E contents but also with the corresponding different meat PUFA proportions, as PUFA are more susceptible to oxidation than MUFA and especially SFA. Unlike the present results, the induction time was clearly lower in perirenal fat of bulls fed grass silage compared to maize silage in the study by Staerfl et al. (2011), but in that study, values of induction time found for grass silage were still higher than those in the present study. This may have resulted from a much higher proportion of more stable SFA typically found in perirenal compared to subcutaneous fat (Wolf et al., 2020).

4.3. Fatty acid profile of the intramuscular fat

The intramuscular FA profile differed among diets, especially with respect to proportions of the $n-3$ FA. This was expected as feeding grassland-derived feeds, naturally rich in C18:3 $n-3$, is known to increase tissue proportions of $n-3$ FA and thus alter the meat FA profile (French et al., 2000; Wood et al., 2003). Consistent with findings by O'Sullivan et al. (2002), meat C18:3 $n-3$ proportion was more pronounced with increasing dietary amounts of grass silage in the present study compared to the maize-silage based diets. In addition, the proportions of the nutritionally valuable long-chain $n-3$ FA C20:5 $n-3$ and C22:6 $n-3$ (Zarate, El Jaber-Vazdekis, Tejera, Perez, & Rodriguez, 2017) increased with increasing dietary grass silage proportion. Compared to our findings, meat lipids from animals fattened on fresh grass (pasture) provided even higher proportions of C18:3 $n-3$ and long chain $n-3$ FA (Razminowicz, Kreuzer, Leuenberger, & Scheeder, 2008). Although no dietary recommendations for humans are defined regarding the ratio of $n-6/n-3$ FA (FAO (Food and Agriculture Organization of the United Nations), 2010), a ratio below 4:1 is considered as beneficial to maintain general health and prevent cardiovascular diseases (Simopoulos, 2002). To achieve a ratio of $n-6/n-3$ FA below that threshold, a grass silage proportion of at least 500 g/kg was needed according to the present results. When increasing the grass silage proportion to 750 g/kg, this ratio can even be lowered to about 2:1. Nevertheless, when CCM was used instead of maize silage, the $n-6/n-3$ FA ratio was increased by 28% (G500_{CCM} vs. G500), indicating that part of the favourable effect of grass silage feeding is lost. Overall, the increase in $n-3$ FA with increasing proportions of grass silage in the present experiment can be considered efficient since the proportions of C18:3 $n-3$ and C20:5 $n-3$ in meat obtained from G750_{CCM} animals increased to an even higher and a similar extent, respectively, as observed in heifers supplemented with partially rumen protected fish oil particularly rich in C20:5 $n-3$ and C22:6 $n-3$ (Wolf et al., 2018). The increase in C22:6 $n-3$ proportion was however smaller than with fish oil supplementation. A daily intake of 250 mg C20:5 $n-3$ and C22:6 $n-3$ is recommended for human nutrition (FAO, 2010). Consuming 100 g of LTL from G750_{CCM} animals per day would provide humans with 14.7 mg of C20:5 $n-3$ and C22:6 $n-3$, thus meeting only about 6% of the recommended intake. However, consuming also adherent adipose tissue would increase intakes.

Besides the $n-3$ FA, conjugated linoleic acids (CLA) are of particular interest. This especially concerns the isomer C18:2 *cis-9*, *trans-11*, the most prevalent CLA found in ruminant-source foods, due to its anticarcinogenic, antiobesogenic, antidiabetic and antihypertensive properties

(reviewed by Koba & Yanagita, 2014). The FA C18:2 *cis*-9, *trans*-11 occurs as an intermediate product of ruminal biohydrogenation of C18:2 *n*-6 and from endogenous synthesis in the tissue from C18:1 *trans*-11 by the Δ^9 -desaturase (Koba & Yanagita, 2014; Scollan et al., 2006). In our study, the proportion of C18:2 *cis*-9, *trans*-11 was higher in G100 than in G750_{CCM}. This can be explained by the about three times higher proportion of C18:2 *n*-6 in the maize-based forages compared to the grass silage, the FA used as substrate for the isomerisation to C18:2 *cis*-9, *trans*-11. Nielsen, Sejrsen, Andersen, Lund, and Straarup (2004) also found higher CLA proportions in milk from dairy cows fed maize silage instead of grass silage. The study of Bu, Wang, Dhiman, and Liu (2007) demonstrated that indeed diets rich in C18:2 *n*-6 compared to diets rich in C18:3 *n*-3 led to higher C18:2 *cis*-9, *trans*-11 proportions in milk, indicating a more intensive biohydrogenation of C18:3 *n*-3 compared to C18:2 *n*-6.

5. Conclusion

Based on the results of the present study, the dietary level of grass silage for fattening bulls can be raised to 500 g/kg diet DM without impairing growth performance, while the slaughter performance was maintained for a diet even containing 750 g/kg grass silage. Thus hypothesis (1) was partly disproven. However, the growth performance was only maintained with 500 g/kg grass silage when complementing this diet with CCM, a particularly energy-dense forage, which confirms hypothesis (2). Although some physicochemical meat quality parameters varied between groups, no clear diet effect was found and neither meat nor sensory quality were adversely affected, thus disproving hypothesis (3). Hypothesis (4) was confirmed as the proportion of *n*-3 FA in meat lipids increased with elevating amounts of grass silage in the diet. Concomitantly, the ratio of *n*-6/*n*-3 FA in the meat was favourably lower compared to a common maize-silage and concentrate-based diet, thereby improving the nutritional quality for the consumers. A certain part of this favourable lowering of the *n*-6/*n*-3 FA ratio was lost when using CCM instead of maize silage. A diet containing 500 g/kg grass silage also seems to provide sufficient rumen-degradable protein for microbial protein synthesis, providing adequate amounts of ruminally available energy for microbial protein synthesis *via* the CCM. Harvesting the grass silage at the optimal development stage and with high conservation quality may further increase the suitability of dietary grass silage for fattening bulls.

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CRedit authorship contribution statement

M. Keller: Conceptualization, Methodology, Formal analysis, Investigation, Data curation, Writing – original draft, Writing – review & editing, Visualization. **M. Kreuzer:** Conceptualization, Resources, Writing – review & editing, Supervision. **B. Reidy:** Conceptualization, Writing – review & editing. **A. Scheurer:** Conceptualization, Writing – review & editing. **B. Guggenbühl:** Methodology, Formal analysis, Resources, Writing – review & editing. **M. Luder:** Methodology, Resources, Writing – review & editing. **J. Frank:** Methodology, Resources, Writing – review & editing. **K. Giller:** Conceptualization, Resources, Methodology, Formal analysis, Investigation, Writing – review & editing, Visualization, Supervision.

Declaration of Competing Interest

The authors declare that there is no conflict of interests.

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