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Effect of the Feed Enzyme Amylase on Growth Performance, Nutrient Digestibility and Meat Quality of Beef Cattle

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Abstract: The objective of this study was to evaluate the effect of an exogenous amylase preparation on fattening bull growth performance, apparent total tract nutrient digestibility and meat quality. For this purpose, a total of 72 fattening bulls with a mean age of 210 days and a mean weight of 260 kg were assigned to six dietary treatment groups, with 12 bulls each for 252 days fattening trail. The control group (1st treatment) fed a ration high in maize product (in average 38% starch) without amylase enzyme, the other five groups fed the control ration supplemented with amylase enzyme in graded levels of 0.5, 1.0, 2.0, 3.0 and 6 ml per kg dry matter intake (DMI) for 2nd, 3rd, 4th, 5th and 6th treatments, respectively. Growth performance parameters (live body weight, feed intake and average daily gain) of bulls in the different treatments was recorded. Titanium dioxide was added in a rate of 0.1% of the total dry matter intake to the concentrate of the control group, the groups fed on 2.0 and 6.0 ml amylase per kg dry matter intake to determine nutrient digestibility. Meat quality in terms of carcass weight, weight of kidney fat, meat colour, meat pH and chemical composition was also determined. The obtained results indicated that supplementation of the enzyme amylase had no effect on feed intake but it increased the live body weight and average daily gain numerically but not significantly. The concentration of amylase enzyme was also clear on live body weight and average daily gain as the low amylase concentration (0.5 ml/kg DMI) showed no effect, whereas high concentration (2.0 or 3.0 ml/kg DMI) are most successful. Amylase supplementation improved the digestibility of organic matter and this improvement restricted mainly in a better digestibility of crude fiber and neutral detergent fiber, while it had no effect on starch digestibility. There were no significant effect of the supplementation of amylase enzyme on meat quality, but meat colour and intramuscular fat numerically showed better values in the treatment groups fed amylase in comparison to the control group. In conclusion, amylase enzyme could be used as a ruminant feed supplement to improve performance and organic matter digestibility as well some improvement in meat quality like colour and intramuscular fat due to addition of amylase enzyme were observed.

Key words: Amylase · Performance · Digestibility · Carcass · Meat quality

INTRODUCTION

Exogenous enzymes have been used extensively to remove anti-nutritional factors from feeds, to increase the digestibility of existing nutrients and to supplement the activity of the endogenous enzymes of poultry [1]. Researchers in the 1960s examined the use of exogenous enzymes in ruminants [2 - 4], but responses were variable. In recent years there has been interest in re-examine the potential use of enzyme in ruminant nutrition. This interest stems from the high cost of livestock production, the availability of new enzyme mixtures with low cost and the potential economic returns to be realized with effective enzyme supplementations [5]. A lot of studies were made in the range of the use of fibrolytic enzymes for dairy cows or growing cattle, but the results of those studies are much differentiated. Besides plant fiber starch is becoming more important in

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rations of high yielding dairy cows or in intensive feeding of fattening cattle. In the rumen starch is degraded to volatile fatty acids, whereas propionic acid the main precursor of gluconeogenesis is produced in a relative high parentage. However, under conditions of intensive feeding (dry matter intake) the passage rate of the digesta is high and the incubation time in rumen is short. Therefore, undegraded starch reach the small intestine [6, 7]. The site of starch digestion alters the nature of the end products of digestion (i.e volatile fatty acids in the rumen and hind gut and glucose in the small intestine) and, in this respect, the efficiency of their metabolic utilization by the ruminant [6]. Also, especially starch of maize kernels and of sorghum show a low ruminal degradability, only about 50 to 60% [8, 9]. Therefore, the amount of undegraded starch increases in the small intestine. But the capacity of the small intestine to digest starch and to absorb glucose is limited [6, 10, 11]. Depending on the components of the ration and under high feeding conditions there is an interest to increase the ruminal degradation of starch and to reduce the overflow of the intestine with starch. Although there is abundant information on the use and mode of action of exogenous fibrolytic enzymes in ruminants [12], the number of studies on exogenous amylase is still small and the exact mechanisms by which amylases might improve digestibility have not been fully explored. Eisenreich [13], shows that the feed enzyme amylas significantly increases the ruminal degradability of maize kernels using the in situ method. Therefore, we plan an in vivo experiment using condition of intensive feeding and a ration of high percentage of maize products. The objective of this study was to investigate the effect of the feed enzyme amylase (Amylase-7b®) on growth performance, nutrients digestibility and meat quality parameters of beef cattle.

MATERIALS AND METHODS

Experimental Design, Animals and Animal Housing: The present work was conducted at the Research Experimental Station of Chair of Animal Nutrition, Center of Life and Food Sciences, Weihenstephan, Technische Universität München, Germany. The care, maintenance and handling of the animals were carried out according to the guidelines of the German laws for animal care. In a total of 72 animals were assigned to six treatment groups with 12 bulls each. The control group (1st treatment) received a ration high in maize products (in average 38% starch) without amylase enzyme the other five groups receiving the control ration in addition to amylase in graded level of 0.5, 1.0, 2.0, 3.0 and 6 ml per kg DMI for 2nd, 3rd, 4th, 5th and 6th treatments, respectively. Amylase enzyme (Amylase-7b®, Novozymes) is declared an Alpha amylase with 240 KNU S/ g and produced from bacillus licheniformis.

All animals were pure-bred Simmental (Deutsches Fleckvieh). The animals were purchased from the cattle market as male calves with an age of 178±20 days and an average live weight of 215±24 kg. They were housed in the fattening pen afterwards. Before the beginning of the experiment they were adapted to the experimental conditions (housing and feeding system) for one month. The bulls had a mean age of 210 days and starting weight of 260 kg. The bulls were kept in pens with fully slatted floors in groups of six animals. Every animal had its own feeding trough which only could be entered by the individual animal (individual feeding system "Calan").

The experimental period extended for about 8.5 months (252 days) and it was divided in to two phases. The 1st phase extended for 168 days and it representing the highest daily weight gain in the main growth period whilst, the 2nd phase extend for 84 days and it representing the medium daily weight gain in the finishing period.

Feeding Ration: All animals had free access to maize silage in both phases of the experiment, while, the concentrate (soybean meal 40%, corn grain 56.3% and minerals and vitamins 3.7%) was fed restricted with 2.2 kg in the first two months, 2.6 kg in the 3rd and 4th month, 3.0 kg in the 5th and 6th month and 3.2 kg in the last two and half months. The increase in concentrate done according to the increasing feed intake of bulls in the course of the trail. The concentrate were mixed with the amylase enzyme weekly. The liquid solution was thinned in water and sprayed on the concentrate during the mixing process. The amount of amylase was continuously adapted to the increasing dry matter intake of the bulls. The amylase concentration per kg dry matter intake was calculated depending on the amylase additive applied to the concentrate and total feed intake. To be certain from the actual concentration and mixing of amylase, every two weeks regular analysis to detect and adjust the concentration of amylase was done.

Measurements

Feeding Experiment: As the experiment extended for 252 days, the bulls were weighted in the morning before feeding on a balance installed in the fattening house at

the beginning of the trail as well as every two weeks. The maize silage and concentrate quantity was weighed daily individually for every animal, the feed refusal every second day. The feed intake was calculated from the difference of these weighs. Thereby, no concentrate residuals were assumed. Every week three samples of the maize silage was drawn and stored deep frozen. The dry matter of maize silage was measured in a pooled sample from 14 days after oven drying at 60°C for 24 hours. Samples from the concentrate were drawn weekly and were mixed for every four weeks. After grinding the dried sample, the contents of crude nutrients (crude protein, ether extract, crude fiber and ash) were determined by the use of Weender analysis [14] in four weeks mixed samples. Starch contents of the maize silage were measured according to Naumann and Bassler [15]. The contents of metabolizable energy (ME) of the maize silage was calculated by the analyzed crude nutrients and their digestibilities. The digestibility values were calculated from the feed value table for ruminant [16]. The content of ME in the concentrate was calculated according to the single components of the concentrate and their energy content [16]. The ME content of the total ration was calculated depending on the relative percentage of maize silage and concentrate in the ration (dry matter basis).

Digestibility Trail: Using titanium dioxide (TiO_2) as a marker, digestibility of the ration was also measured with the bull during the feeding experiment. For this purpose all animals in the control group, the 4th treatment group (2.0 ml amylase per kg DMI) and the 6th treatment group (6.0 ml amylase per kg DMI) were used. In total 36 animals (12 bulls per treatment) were part of this digestibility trail. The experiment was divided into a 14 day preparatory period and a five days collecting period. Titanium dioxide was mixed into the concentrate at a rate of 0.1% from the total DMI. Maize silage and concentrate were fed in a constant amount during the digestibility trail. Therefore, the bulls individually received as much feed as they consumed without any residual. During the collecting period faecal matter was taken rectally in the morning before feeding. The faecal sample was homogenized and 200 g were weighed into a container for each animal and stored deep frozen. At the end of the digestibility trail, the entire faecal sample per animal was freeze dried and ground. The Weender analysis [14] was used to determine the crude nutrients in the faeces and the feed. In faeces and feed the evaluation of starch has been carried out using the enzymatic method of Seidler *et al*. [17]. For evaluation of NDF and ADF content, the method of Goering and Van Soest [18] was used. The TiO_2 content in the concentrate and the faeces was determined according to the method of Brandt and Allam [19] and Brandt *et al*.[20].

Meat Quality

Slaughter Procedure: The bulls were fasted for 18 h before slaughtering at a commercial abattoir. The carcass were trimmed according to the EU legislation. Carcass weight, weight of kidney fat, classification of carcass (dressing percentage, EUROP) and fat category was determined. Carcasses were split and cooled for 24 h at 4°C. The best ribs (9th to 11th) were according to method of Hankins and Howe [21], removed from the left side of the carcasses and kept at +4°C during the transportation to the Bavarian Institute of the Animal breeding, Grub, Munich, Germany. All meat samples were stored at +4°C for further meat quality measurement. Samples from the longissimus dorsi (LD) muscle were stored at -20 for crude nutrient and intramuscular fat analysis according to the method of AOAC [22].

Meat Quality Evaluation: Meat pH was measured in the LD muscle 48 h after slaughter. The Minolta reflexion photometer was used to measure meat colour (L* = lightness; a* = redness; b* = yellowness) according to CIE [23]. Measurements of colour values were carried out at five spots of the LD muscle. The LD muscle were cut into three steaks at the height of the 9th, 10th and 11th rib. Each LD steak of the 9th rib was trimmed to remove residual adipose tissue and connective tissue for subsequent blending. From the homogenised meat direct measure of the intramuscular fat (IMF) were performed using the near infrared reflectance method. The 10th rib steaks (3 cm thickness) were used for the determination of shear force values, aging and grilling losses percentages according to Orellana *et al.* [24]. Each of these steaks was weighed, vacuum packaged and aged at +2°C for 13 days. The steaks were weighed again and the ageing loss percentage was calculated. Subsequently, the steaks were heated until a core temperature of 65°C, grilling time was recorded and the grilled samples were cooled to room temperature and weighed again to calculate the grilling loss percentage. Shear force was measured in the grilled steaks.

Statistics Analysis: All data were statistically evaluated by use the SAS program [25]. Separate evaluations were carried out for the respective 14 day weighing and measuring periods as well as for the entire period of the trail. The mean values of the treatments with the standard deviation of the single values are specified in the tables.

RESULTS

Characteristics of the Feed: Chemical composition of maize silage in four weeks period are illustrated in Table 1. The mean dry matter content of 41.7% for the maize silage is consistent with a relatively late harvest date. Further on, during the harvest the maize plant were cut about 40 cm above the bottom. Therefore, the maize silage had a clear cob emphasis. The mean crude fiber content was very low with 16.8% . However, the starch content was very high with an average value of nearly 41%. Therefore, the corresponding content of ME was high 11.17 MJ/kg DM. Chemical composition of concentrate as a mean over the experimental period are shown in Table 2. The concentrate consisted approximately one half of each of soybean meal and corn grain. The concentrate had a mean crude protein and starch content of 27.0% and 36.6%, respectively and had an average content of 12.24 ME/kg DM.

Effect of Feeding Amylase Enzyme on Bull Growth Performance

Live Weight Gain: The results of the average live weight of bulls are presented in Table 4. The animals have had a relatively homogeneous mean live weight of 260±24.7 kg at the start of the experiment. The mean live weights of animals at the end of the 1st phase and the 2nd phase were 532±36.4 and 631±41.5 kg, respectively. At the end of the 1st phase the bulls fed on 2 ml amylase per kg DMI showed an 18 kg higher average live weight of 543 kg in comparison to 525 kg live weight reached by the control group. The bull in treatment groups received 1 ml, 3 ml and 6 ml amylase per kg DMI had a final weight of 534 kg, 531 kg and 536 kg, respectively at the end of 1st phase, which were nearly 10 kg higher than the control group. Only bulls in treatment group which received 0.5 ml amylase per kg DMI showed no difference with the control. Due to the high variance between the individual animals in each group, no statistical significant differences were seen.

At the end of 2nd phase, the live weight of bulls received 1.0 ml A/kg DMI (638 kg) and 2.0 ml A/kg DMI (637 kg) showed 14 kg and 13 kg respectively, higher than the control group (624 kg). Whereas, the bull in treatment group received 3.0 ml A/kg DMI (633 kg) and 6.0 ml A/kg DMI (630 kg) showed 9 kg and 6 kg respectively, higher live weight in comparison to control group. Again the treatment received 0.5 ml A/kg DMI showed no effect of amylase supplementation on live body weight (622 kg).

Feed Intake: The results of the feed intake of the bull in the different treatments are presented in Table 4. The animals showed a average feed intake of 7.58±0.35 kg from the start of the experiment to the end of 1st phase. In the 2nd phase feed intake of the animals increased to an average value of 8.97±0.48 kg. Accordingly from the start to the end of the trail the bulls had an average feed intake of 8.04±0.36 g. The feed intake of the bulls remained relatively unaffected by amylase enzyme supplementation.

The average feed intake (kg DM) and energy intake (MJ ME) was subdivided into the intake of maize silage and concentrate (Table 3). These data clarify that feed and energy intake nearly describe a plateau in the last four months which might affect ADG. In total about two-thirds of the energy comes from maize silage and one-third comes from concentrate.

Average Daily Gain: The results of the average daily gain of the bull in the different treatments are presented in Table 4. The animals showed an average daily weight gain (ADG) of 1616±137 g from the start of the experiment to the end of 1st phase. In the 2nd phase the animals showed ADG of 1178±252 g. Accordingly from the start to the end of the trail the bulls showed average weight gain of 1470±129 g. In comparison to data reached under practical farm condition the measured daily weigh gain represents very high growth rates (+200 g). Nevertheless, in the 1st phase the ADG of bulls in treatments received 2.0 ml A/kg DMI (1627 g) and 3.0 ml A/kg DMI (1651 g) showed 96 g and 72 g, respectively higher than that of control group (1579 g). The average daily weight gain in groups received 1.0 ml A/kg DMI (1624 g) and 6.0 ml A/kg DMI (1625 g) showed a smaller increase of 45 g and 48 g compared to the control group. In the 2nd the ADG of all groups were considerably lower than in the 2nd phase. Group received 1.0 ml A/kg DMI showed only appreciable increases in ADG (+55 g) with mean daily weight gain of 1236 g compared to the control (1181 g). Over the whole experimental period the bulls in the treatment groups received 1.0, 2.0 and 3 ml A/kg DMI showed numerically higher ADG (+50 g) in comparison to the control bulls, whereas the bulls in group with 6.0 ml A/kg DMI showed little and the group with 0.5 ml A/kg DMI showed no effect for amylase supplementation on ADG.

Table 1: Chemical composition of maize silage

Period	DM (%)	CP%	EE%	Ash	CF%	Starch%	ME/ kg DM (MJ)
		----------	----------	(% of DM)	----------	----------	
1st month	39.8	7.98	3.29	3.10	16.0	43.6	11.16
2nd month	40.2	7.92	3.09	3.13	17.2	36.8	11.12
3rd month	39.7	8.49	3.16	3.05	17.8	41.3	11.15
4th month	40.9	8.40	3.11	2.94	17.2	35.3	11.16
5 th month	41.4	7.91	3.62	3.02	17.5	38.8	11.20
6 th month	41.2	7.81	3.48	2.61	16.6	42.8	11.23
7 th month	43.4	8.03	3.47	3.05	17.8	39.6	11.18
8 th month	44.3	8.11	3.12	2.81	16.1	41.9	11.17
9 th month	44.4	6.74	3.15	2.57	15.4	45.1	11.18
Mean	41.7	7.93	3.28	2.92	16.8	40.6	11.17

Table 2: Chemical composition of concentrate in the different treatments

Treatment	DM (%)	CP%	EE%	Ash	CF%	Starch%	ME/kg DM (MJ)
		----------	----------	(% of DM)	----------	----------	
Control	89.3	27.0	2.51	7.81	5.05	36.7	12.36
0.5 ml A/kg DM I	89.2	27.0	2.77	7.61	5.40	36.5	12.39
1.0 ml A/kg DMI	89.0	27.8	2.89	7.88	5.45	36.3	12.34
2.0 ml A/kg DMI	89.1	28.0	2.81	7.89	5.11	36.5	12.39
3.0 ml A/kg DMI	89.3	27.6	2.61	7.95	5.16	36.6	12.28
6.0 ml A/kg DMI	89.6	27.5	2.61	7.70	5.27	36.0	12.37

DM, dry matter; CP, crude protein; EE, Ether extract; CF, crude fiber; ME, Metabolizable energy; MJ, mega joule

Table 3: Average feed and energy intake of animals

Treatments	Feed intake	Months 1 and 2		Months 3 and 4		Months 5 and 6		Months 7 and 8	
		----------		----------		----------		----------	
		Kg DM	ME (MJ)	Kg DM	ME (MJ)	Kg DM	ME (MJ)	Kg DM	ME (MJ)
Control	Total	6.37	73.7	7.95	91.7	8.72	100.7	8.97	103.7
	MS	4.43	49.6	5.66	63.4	6.10	68.1	6.18	69.3
	Conc.	1.94	24.0	2.29	28.3	2.64	32.6	2.79	34.5
0.5 ml A/kg DMI	Total	6.2	71.8	7.29	88.8	8.41	97.21	8.64	100.1
	MS	4.26	47.7	5.40	60.5	5.77	64.6	5.85	65.6
	Conc.	1.94	24.0	2.29	28.3	2.64	32.6	2.79	34.5
1.0 ml A/kg DM I	Total	6.28	72.7	7.87	90.8	8.96	103.4	9.23	106.6
	MS	4.34	48.6	5.58	62.5	6.32	70.8	6.44	72.1
	Conc.	1.94	24.0	2.29	28.3	2.64	32.6	2.79	34.5
2.0 ml A/kg DMI	Total	6.38	73.8	7.98	92.1	8.87	102.3	9.14	105.6
	MS	4.44	49.7	5.69	63.8	6.23	69.75	6.35	71.1
	Conc.	1.94	24.0	2.29	28.3	2.64	32.6	2.79	34.5
3.0 ml A/kg DMI	Total	6.44	74.5	7.95	91.7	8.61	99.5	9.13	105.5
	MS	4.50	50.4	5.66	63.4	5.97	66.9	6.34	71.0
	Conc.	1.94	24.0	2.29	28.3	2.64	32.6	2.79	34.48
6.0 ml A/kg DMI	Total	6.33	73.2	7.89	91.0	8.63	99.7	8.77	101.5
	MS	4.39	49.2	5.6.0	62.7	5.99	67.1	5.98	67.0
	Conc.	1.94	24.0	2.29	28.3	2.64	32.6	2.79	34.5
Mean	Total	6.33	73.2	7.89	91.0	8.70	100.5	8.98	103.8
	MS	4.39	49.2	5.60	62.7	6.06	67.9	6.19	69.3
	Conc.	1.94	24.0	2.29	28.3	2.64	32.6	2.79	34.5

MS, Maize silage; Conc. Concentrate; DMI, dry matter intake

Table 4: Average live body weight (kg), feed intake (kg) and average daily gain (g) of the bulls during the first and second phase of feeding

		Amylase content per kg DMI of the ration					
Parameter	Period	0	0.5 ml	1.0 ml	2.0 ml	3.0 ml	6.0 ml
Live body Weight (kg)	Starting body weight	259±20.1	262±26.2	261±26.8	262±22.7	254±32.7	263±22.4
	Phase 1	525±40.9	521±44.6	534±32.5	525±23.1	531±45.6	536±29.5
	Phase 2	624±53.8	622±39.1	638±45.4	637±34.6	633±44.1	633±31.8
Feed intake (kg)	Phase 1	7.62±0.26	7.39±0.56	7.66±0.33	7.67±0.32	7.58±0.32	7.56±0.29
	Phase 2	8.96±0.45	8.62±0.54	9.18±0.41	9.15±0.47	9.05±0.47	8.82±0.40
	Total	8.07±0.30	7.80±0.52	8.17±0.33	8.16±0.32	8.16±0.30	7.98±0.30
Average daily gain (g)	Phase 1	1579±171	1538±146	1624±140	1675±75	1651±122	1627±128
	Phase 2	1181±316	1206±208	1236±214	1120±316	1207±207	1211±138
	Total	1446±164	1428±78	1495±152	1495±152	1503±98	1470±124

Table 5: Apparent total tract digestibility of organic matter, fiber fractions (CF, NDF and ADF)

	Digestibility%				
Treatment	OM	CF	NDF	ADF	Starch
Control	75.0±4.86	55.4±5.84	60.5±4.48	57.0±5.37	96.0±1.38
2.0 ml A/kg DMI	77.4±3.54	58.5±5.53	62.9±5.54	57.7±5.93	96.4±0.90
6.0 ml A/kg DMI	77.6±3.79	60.2±5.88	64.8±5.79	57.2±4.64	96.4±1.11

DM, dry matter; CF, crude fiber; NDF, neutral detergent fiber; ADF, acid detergent fiber

Table 6: Carcass weight, kidney fat weight, classification of carcass by EUROP (dressing percentage) and fat category (adipose)

	Amylase content per kg DM of the ration					
Parameter	0	0.5 ml	1.0 ml	2.0 ml	3.0 ml	6.0 ml
Carcass weight (kg)	361.5±34.7	355.1±25.7	365.5±27.5	362.7±21.6	357.9±30.1	358.6±21.0
Kidney fat (Kg)	12.1±3.20	11.5±3.20	11.4±3.40	12.5±3.10	11.5±2.60	10.5±3.10
Classification						
EUROP	3.70±0.48	3.36±0.67	3.75±0.45	3.75±0.45	3.75±0.45	3.67±0.49
Fat category	2.50±0.53	2.55±0.52	2.67±0.49	2.50±0.52	2.67±0.49	2.50±0.52

*E=5, U=4, R=3, O=, P=1

**1(minor adipose) – 4 (highly adipose)

Table 7: Meat colour classification

	Amylase content per kg DM of the ration					
Parameter	0	0.5 ml	1.0 ml	2.0 ml	3.0 ml	6.0 ml
L*	35.6±3.10	34.9±3.00	36.5±1.20	36.6±1.80	37.1±3.00	37.2±2.10
a*	10.4±2.10	10.6±1.20	10.8±1.40	11.4±1.00	11.6±1.60	11.4±0.80
b*	1.40±2.52	2.06±0.87	2.30±1.25	1.99±1.42	2.78±1.63	4.03±1.47

L* = lightness; a* = redness; b* = yellowness

Table 8: Ultimate pH value (post ageing), shear force value (tenderness), ageing and grilling losses of meat

	Amylase content per kg DM of the ration					
Parameter	0	0.5 ml	1.0 ml	2.0 ml	3.0 ml	6.0 ml
pH value (post ageing)	5.76±0.31	5.80±0.22	5.68±0.14	5.60±0.08	5.62±0.09	5.60±0.17
Shear force (kg cm)	3.55±0.66	3.73±0.61	3.67±0.79	3.74±1.45	4.02±1.40	3.09±0.52
Shear energy (J)	0.31±0.04	0.32±0.04	0.31±0.07	0.32±0.10	0.34±0.10	0.26±0.04
Ageing loss (%)	2.78±0.88	2.85±0.70	3.31±0.82	3.43±0.77	3.14±0.58	3.19±0.99
Grilling loss (%)	24.3±4.30	25.3±3.10	26.4±2.10	27.5±2.50	26.5±3.10	26.5±2.80

Table 9: Chemical composition of longissimus dorsi (LD) muscle

	Amylase content per kg DM of the ration					
Parameter	0	0.5 ml	1.0 ml	2.0 ml	3.0 ml	6.0 ml
Water (%)	73.4±0.05	73.9±0.90	73.8±0.60	73.0±0.70	73.2±0.40	73.4±0.50
Crude protein (%)	1.08±0.03	1.07±0.01	1.08±0.02	1.08±0.03	1.07±0.02	1.07±0.02
Intramuscular fat (%)	2.59±0.33	2.21±0.76	2.12±0.48	2.92±0.64	2.85±0.54	2.71±0.74
Ash (%)	23.0±0.90	22.6±0.70	22.7±0.80	23.2±0.90	22.9±1.00	22.3±0.80

Apparent Total Tract Digestibility: The values of digestibility of OM, CF, NDF, ADF and starch of the three treatments (control, treatment groups received 2.0 and 6.0 ml A/kg DMI) are shown in Table 5. Depending on the high quality of all rations the digestibility of OM is high. However the addition of amylase enzyme affected the rate of digestion. The digestibility of OM raised from 75.0% (control group) to 77.4% (+2.4%) in group received 2.0 ml A/kg DMI and in 77.8% (+2.8%) in group received 6.0 ml A/kg DMI. This improvement in digestibility was most obvious regarding to the fibrous fraction of feed. Digestibility of crude fiber of treatment group received 2.0 ml A/kg DMI (58.5%) and group received 6.0 ml A/kg DMI (60.2%) was higher than the control (55.4%) with 3.1% and 4.2%, respectively. The value of NDF digestibility of both groups was (62.9 and 64.8%) with 2.4 and 3.9% higher in digestibility in comparison to the control group (60.5%). In total the level of digestibility of the ADF was low about 58% on the average of all treatment groups. With a value of 96%, starch digestion was nearly completely in all groups. However, this might be seen in context with the high level of feeding which affects the digestibility in a depressing way. The supplementation of amylase tended to result in favourable effects on the digestibilities of OM, CF and NDF compared to the control group, where the digestibilities of ADF and starch showed no effect relative to amylase supplementation.

Carcass and Meat Quality: Carcass weight, kidney fat weight, classification of carcass by EUROP (dressing percentage) and fat category (adipose) of the animals by treatment groups are shown in Table 6. All data are in normal range and addition of amylase showed no effect on all parameters. Classification shows high values beside treatment group received 0.5 ml A/kg DMI, which was in tendency lower (3.36). Surprisingly kidney fat and fat category was not increased even by the high feeding intensity and growth rate.

The results of the meat colour of LD muscle (lightness, redness and yellowish) measurement are presented in Table 7. Treatment groups fed 2.0, 3.0 and 6.0 ml A/kg DMI numerically gives higher values for redness, which again affects the lightness. A more "red" beef is very positive quality marker for selling fresh meat.

Meat ultimate pH value (post ageing), shear force value (tenderness), ageing and grilling losses of the different treatments are presented in Table 8. Supplementation of amylase enzyme didn't affect all of those parameters. Again all data show a good quality, especially the values of shear force are in the range of "tender" meat which normally might be a problem with bulls meat.

The chemical composition of LD muscle of the animal in the different groups are shown in Table 9. Focused on the intramuscular fat values, the control group and the treatment fed 0.5 and 1.0 ml A/kg DMI have a mean value of 2.3%, whereas the groups fed 2.0, 3.0 and 6.0 ml A/kg DMI have a mean value of 2.8%. Intramuscular fat in the range of 2.6 – 2.9% are preferred, whereas values of < 2.5% are typically for bulls of the breed Simmental.

DISCUSSION

This study was designed to assess the effect of amylase enzyme on growth performance at high starch ration. Therefore, the ration mainly consisted of maize products like maize silage (fed *ad-libtum*) and maize grain (about 50% of the used concentrate). The starch content of the ration was very high (in average 38% from DM) and the crude fiber was in a lower range (in average 13/14%). The mean of crude protein content was nearly 12%. This ration expressed a high feeding intensity. Addition of amylase enzyme didn't affect animal feed intake. These results are in line with the previous study of Noziere *et al.* [26], as he found that amylase had no effect on feed intake. In the current study the feed intake quickly raised with increasing animal live weight. But very soon the feed intake showed a plateau with no further increase. Feed and energy intake nearly describe a plateau in the last four months which might affect ADG. This corresponding to a typical curve of feed intake in an intensive feeding system. The average feed intake

(kg DM) and energy intake (MJ ME) was subdivided into the intake of maize silage and concentrate. In total about two-thirds of the energy comes from maize silage and one-third comes from concentrate. The overall mean of daily DMI was 8.0 kg per animal the energy intake was in average of 96 MJ ME per animal per day.

The live weight and the ADG of the different feeding groups are in a good relation to the feed intake. The different groups showed a very high growth rate and a typical growth curve for an intensive feeding system. Beside this high growth rate, addition of amylase increases the ADG especially in the 1st phase from 1579 g (control group) up to 1675 g with addition of 2.0 ml amylase per kg DMI. However the standard deviation between the animals was high and there were no statistical differences. Further on, there was a clear effect on the increasing amylase concentration. The addition of 0.5 ml amylase per kg DMI showed no effect, whereas addition of 2.0 or 3.0 ml amylase per kg DMI were most successful. These results are in agree with Tricarico *et al.* [27] he reported that an *A. oryzae* extract containing α-amylase activity quadratically increased ADG of beef cattle when included in either a cracked corn or high-moisture corn and corn silage diet but had no effect when included with alfalfa hay, cotton seed hulls, or steam-flaked corn.

The effect of amylase enzyme on nutrients digestibility showed increase in organic matter digestibility in comparison to control. Further on, the increase in organic matter digestibility was related to increased crude fiber and NDF digestibility. The starch digestibility was nearly completely digestible 96%. Therefore starch digestibility didn't affected with addition of amylase. But this digestibility gives no answer, in which part of the gastrointestinal tract (rumen or intestine or hindgut) starch was digested. The place of degradation can affect the energy value of starch too. The site of starch digestion alters the nature of the end products of digestion (i.e volatile fatty acids in the rumen and hind gut and glucose in the small intestine) and, in this respect, the efficiency of their metabolic utilization by the ruminant [6]. In agree with the previous data, several studies have demonstrated that exogenous amylase preparations resistant to ruminal degradation are able to improve OM digestibility in dairy cows [28-30] or beef steers [31]. This increased OM digestibility was associated, in some cases, with improved NDF digestibility, whereas no effect on starch digestibility [29, 30, 32] or on true digestibility of OM in the rumen [28] was found. Noziere *et al.* [26] despite the improved ruminal starch digestion with amylase supplementation, he found that total-tract starch digestion was not modified, reflecting the ability of the intestine to digest starch escaping ruminal digestion, which can be up to 3 g/d per kg of BW [6, 11]. Previous studies reported a numerical increase in total-tract digestibility of starch with amylase-supplemented but it never exceeded 1.7% and was not always observed [33]. According to Sami *et al*. [34], all parameters of carcass and meat quality were in good relation to high slandered of fattening bulls especially for bread Simmental. There were no significant effects for amylase addition on meat quality. But some important parameters of meat quality like colour or intramuscular fat showed better values in the treatment groups with amylase in comparison to the control group or a low addition of amylase. Meat colour plays an important role in a consumer's purchase decision and may be influenced by a number of pre- and post-slaughter factors [35]. Meat lightness is often inversely correlated to haem iron content, which increases with age [36]. A part of the lightness variation can also be explained by changes in ultimate pH and intramuscular fat content [37]. The increase of pH measured 48 h post mortem caused the deterioration of colour parameters in meat from different cattle categories [38]. Ageing time is one of the most important factors influencing most of the sensory properties, especially tenderness [39]. Intramuscular fat in the range of 2.6 – 2.9% are preferred, whereas values of < 2.5% are typically for bulls of the breed Simmental [34].

CONCLUSION

The use of exogenous enzymes amylase in fattening bull is still an emerging technology. The exogenous amylase used in this study improved ADG and body weight of bulls at the intensive feeding system and this may be attributed to the improvement in nutrients digestibility in form of OM, CF and NDF, but not starch. Some improvement in meat quality like colour and intramuscular fat due to addition of amylase enzyme were observed. Further research on high-producing animals is necessary to assess the usefulness of this exogenous amylase to improve starch digestion when ruminal conditions are less favourable.

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