A note on distribution of lipolytic activity in the digestive tract of veal calves^{*}

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ABSTRACT

Six Holstein calves, 3 to 4 weeks of age were fed a milk replacer and a starter concentrate. Calves were slaughtered after 105 days, without fasting. Contents of the rumen, abomasum, duodenum, jejunum + ileum, caecum and colon were weighed, the pH was measured and the rate of hydrolysis of emulsified tributyrin determined. In all calves, the highest and the lowest specific lipolytic activity (per g digesta) were found in the duodenum and abomasum, respectively. The ruminal lipolytic activity was rather low, but thank to its great volume the rumen contained almost one half of the total lipolytic activity present in the digestive tract. The reflex of oesophagal groove closure, however, restricts the potential role of the rumen in digestion of dietary fat to hydrolysis of lipids present in the solid feed. In pooled samples of digesta from the rumen, abomasum, duodenum and jejunum/ileum the highest lipolytic activity was observed at pH of 5.5, 2.5, 8.5 and 9.0, respectively. There was no clear pH optimum for hydrolysis of tributyrin by the caecal/colonic contents. It can be concluded that lipolytic enzymes are present in all sections of the calf digestive tract, but individual digestive segments differ substantially in specific and total lipolytic activity, and in nutritional significance of these activities for the animal.

KEY WORDS: veal calves, digestion, lipolysis, fat

INTRODUCTION

Fat covers a major part of the calf energy requirement, thus digestion of lipids in preruminant calves deserves steady attention. Fat digestibility in young calves is generally high but may be decreased if inexpensive vegetable fats were added to

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skimmed milk to replace the milk fat in the diet. Digestibility of fat depends on its fatty acid profile, being lower in fats containing saturated long-chain fatty acids, and high in fats with unsaturated and/or short-chain fatty acids (Roy and Stobo, 1975). Fatty acids are released from their ester bonds by lipases, which represent a large family of hydrolases, ubiquitous throughout living organisms. A comprehensive review on lipid digestion in young ruminants was published by Noble (1980). In calves previous studies worth attention were concerned with lipase present in saliva (Grosskopf, 1965; Edwards-Webb and Thompson, 1977, 1978; Toothill and Thompson, 1977), abomasum (Guilloteau et al., 1983, 1985), and pancreatic juice (Edwards-Webb and Thompson, 1977; Toothill and Thompson, 1977; Guilloteau et al., 1983, 1985). Villeneuve et al. (1996) investigated pregastric lipases secreted by the lingual and pharyngeal tissues of young ruminants. Šimůnek et al. (1995) published a paper on ontogenesis of hydrolytic activities in the gastrointestinal tract of young goats. Activities of lipases were very variable and rather low in almost all samples examined. We are not aware of a comparative study dealing with distribution of lipolytic activity in individual segments of the digestive tract of veal calves. Thus, we measured the lipolytic activity in samples of digesta taken from the rumen, abomasum, small intestine and large intestine of 4 months-old calves fed a milk replacer and a starter concentrate. The effect of pH on the lipolytic activity was also investigated.

MATERIAL AND METHODS

Animals and diets

Six Holstein bulls, 3 to 4 weeks of age at the start of the experiment, were fed a milk replacer Telasan V (Bodit Tachov, Czech Republic) and a starter concentrate Telstar (Zea Sedmihorky, Czech Republic). The composition of feeds is presented in Table 1. Calves received the milk replacer twice a day at 0.4 kg in 3 l of water.

Nutrients	Milk replacer ¹	Starter concentrate ²	
Dry matter	930	860	
Crude protein	220	200	
Fat	190	29	
Fibre	13	42	
Ash	70	62	

¹Telasan V contained skimmed milk, plant oils, oilseeds, yeast, soyabean meal, cereal products, vitamin and mineral supplements

²Telstar contained cereals, cereal by-products, oilseed cake, by-products of the sugar industry, antioxidant, vitamin and mineral supplements

Starter was available *ad libitum* and its consumption was measured. Animals were slaughtered after 105 days. Feed was available up to the slaughter.

Sampling

The abdominal cavity was immediately opened, the digestive tract was applied to separate rumen, abomasum, small intestine and large intestine (including the caecum). The contents of the rumen, abomasum, the first sixth of the small intestine (presumably the duodenum), the remaining part of the small intestine (presumably the jejunum and ileum), and the large intestine were removed, weighed and pH was measured. Digesta samples were taken and stored at -70°C until analysis.

Lipolytic activity assay

Three methods of lipase activity measurement were compared. A method based on determination of oleic acid released from emulsified 4.5 mM triolein (Fluka, cat.no. 62314) was not suitable for measurement of low lipolytic activities in the rumen and abomasal contents. Lipase activity measurement using the reflectometer RQflex plus[®] (Merck KGaA, local supplier Merck Ltd., Říčany, Czech Republic) was rapid, but results were given in relative units only. Thus, the lipolytic activity was measured by the modified method of Bier (1955). Samples of digesta (1 g) were incubated with 0.2 M citrate/0.4 M phosphate buffer (9 ml) and emulsified tributyrin (10 ml) at pH corresponding to the respective parts of the digestive tract. The emulsion was prepared by sonication from tributyrin (10 ml), Tween 80 (1 ml) and water (100 ml). The reaction mixture was incubated (37°C, 1 h) and then centrifugated. Liberated butyrate was determined by titration, after steam distillation in the Markham apparatus. Specific activity of lipases was expressed as mmol butyrate liberated per 1 g of digesta in 1 h. Total activity was calculated as the product of specific activity and weight of digesta in individual segments. It was assumed that duodenal content represented 15% of the small intestinal digesta weight. To assess the effect of pH on lipolytic activity, mixed samples were prepared and pH of the reaction mixture was adjusted to a desirable value (4.0 to 6.5, 2.2 to 4.5, 5.0 to 10.5, and 5.0 to 9.0 for rumen, abomasal, small intestinal and large intestinal contents, respectively).

RESULTS AND DISCUSSION

Calves consumed milk replacer and starter concentrate at 84 and 202 ± 34 kg, respectively, and increased the average weight from 45.4 to 167.0 kg in the course of the experiment. Table 2 presents weights of digesta, pH, specific and

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Rumen	Abomasum	Duodenum	Jejunum +	Colon +
			ileum	caecum
11.94 ± 2.80	1.80 ± 0.69	0.47 ± 0.07	2.67 ± 0.38	1.54 ± 0.48
5.42 ± 0.13	3.08 ± 0.38	6.37 ± 0.62	7.16 ± 0.24	6.61 ± 0.16
0.17 ± 0.06	0.14 ± 0.14	1.30 ± 0.55	0.35 ± 0.19	0.31 ± 0.14
2.03 ± 0.87	0.25 ± 0.33	0.61 ± 0.24	0.93 ± 0.46	0.48 ± 0.33
	$11.94 \pm 2.80 \\ 5.42 \pm 0.13 \\ 0.17 \pm 0.06$	$\begin{array}{c} 11.94 \pm 2.80 \\ 5.42 \pm 0.13 \\ 0.17 \pm 0.06 \end{array} \begin{array}{c} 1.80 \pm 0.69 \\ 3.08 \pm 0.38 \\ 0.14 \pm 0.14 \end{array}$	$\begin{array}{cccccccccccccccccccccccccccccccccccc$	RumenAbomasumDuodenum i_{ileum} 11.94 ± 2.80 1.80 ± 0.69 0.47 ± 0.07 2.67 ± 0.38 5.42 ± 0.13 3.08 ± 0.38 6.37 ± 0.62 7.16 ± 0.24 0.17 ± 0.06 0.14 ± 0.14 1.30 ± 0.55 0.35 ± 0.19

Table 2. Weight of digesta, pH, specific and total lipolytic activity in segments of the digestive tract of veal calves fed milk replacer and starter concentrate

mean values \pm SD

¹ expressed as mmol butyrate liberated from tributyrin/h per g digesta

² expressed as mol butyrate liberated from tributyrin/h per segment

total lipolytic activity in segments of the digestive tract. Specific lipolytic activity (per g digesta) was higher in the duodenum than in other digestive segments. The rumen, however, thank to its greater volume contained almost one half of the total lipolytic activity of the whole digestive tract. The data in Table 2 illustrate a high potential role of the rumen in hydrolysis of dietary lipids. However, during drinking the rumen is by-passed due to the reflex of oesophagal groove closure (Guilhermet et al., 1976). Thus, it is obvious that the role of the rumen in fat digestion was limited to lipids present in the solid feed. The lowest specific lipolytic activity was that of abomasal contents, in spite of the fact that tributyrin is the ideal substrate for lipases which hydrolyse fat in the abomasums of milk-fed ruminants (Noble, 1980).

The pH optima of lipases in individual digestive segments were greatly different (Figure 1). The highest activity of lipases towards tributyrin in pooled samples of digesta from the rumen, abomasum, duodenum and jejunum/ileum was observed at pH of 5.5, 2.5, 8.5 and 9.0, respectively. The rate of hydrolysis of tributyrin in the rumen contents of grazing cows reached a maximum at pH 7.0 (Faruque et al., 1974), which is a value higher than pH 5.5 observed in the present experiment. On

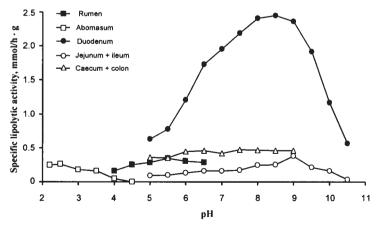


Figure 1. pH-activity curves for the lipolytic activity in sections of the digestive tract of veal calves

the other hand, the pH optimum for hydrolysis of tributyrin in the small intestine (9.0) agrees with the pH optimum reported by Shahani et al. (1976) for bovine pancreatic lipase (8.8). There was no clear pH optimum for hydrolysis of tributyrin by the caecal/ colonic contents. The pH optima of lipases in the rumen and abomasum corresponded to digesta pH in these organs. Lipids in the small intestine, however, were hydrolysed at pH values which were about two pH units below the optimal level. In spite of it, the small intestinal lipolytic activity seems to be high enough to hydrolyse dietary fat completely. Consequently, the role of the large intestine may be limited to hydrolysis of lipids of desquamated epithelial and lysed bacterial cells. As various microbial lipases participate in this process, no distinct pH optimum was obvious.

It can be concluded that lipolytic enzymes are present in all sections of the calf digestive tract, but individual digestive segments differ substantially in specific and total lipolytic activity, and in nutritional significance of these activities for the animal.

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