WEIGHTS OF DIGESTIVE ORGANS, CAECAL METABOLITES AND FERMENTATION STOICHIOMETRY IN COYPUS AND RABBITS

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ABSTRACT

Six coypus and six rabbits fed the same granulated feed *ad libitum* were slaughtered, digestive organs weighed and their contents analyzed. The caecum was the largest digestive organ in both animal species. Its weight averaged 170 g in coypus and 157 g in rabbits (4.1 and 5.5% of the total body weight, respectively). In rabbits, the weight of stomach was greater and that of the small intestine smaller than in coypus (135 and 89 g *vs* 85 and 111 g). Gastric acidity and caecal and colonic dry matter concentrations were significantly higher in rabbits. Total volatile fatty acids (VFA) and ammonia concentrations in the caecal contents of coypus and rabbits were similar (100.1 and 23.3 µmol/g in coypus and 104.8 and 25.5 µmol/g in rabbits, respectively). Molar percentages of acetate and propionate, however, were significantly higher and percentage of butyrate lower in caecal VFA of coypus than in rabbits. The caecal contents were diluted with buffer and incubated anaerobically in order to determine the caecal fermentation pattern. Caecal microorganisms of coypus produced more propionate and methane, and less butyrate and valerate than caecal microbes of rabbits. Thus, different major hydrogen sinks exist in the coypu and rabbit caecum. In conclusion, there are both differences and similarities in the digestion pattern of coypus and rabbits. Caecal fermentation pattern in these herbivore species differed more than other parameters investigated.

Key words: coypu, rabbit, digestion, caecum, fermentation.

INTRODUCTION

Coypus (*Myocastor coypus*) and rabbits are medium-sized herbivore animals with similar morphological features of the digestive tract. In both species the caecum is the primary site of digesta retention and microbial fermentation. Comparative nutritional trials with coypus and rabbits are scarce. HULLÁR *et al*. (1992ab) found that coypus fibre requirement and the significance of caecotrophy were lower, probably due to continuous availability of low-fibre parts of aqueous plants in the coypu´s natural environment. A high-fibre feedstuff (lucerne) was digested less efficiently in coypus than in rabbits, but in

case of feedstuffs low in fibre (maize, wheat) or containing the fibre in an easily digestible form (wheat bran) no difference in digestibility coefficients was observed. Caecal microorganisms of coypus produced more propionate and methane, and less butyrate than caecal microorganisms of rabbits (MAROUNEK *et al*., 1999).
This study has been aimed at extending our knowledge on comparative digestive physiology of herbivores. Thus, weights of digestive organs, acidity and dry matter (DM) concentration of their contents, and concentration of caecal metabolites were determined in coypus and rabbits fed the same diet *ad libitum*, as well as the stoichiometry of fermentation in cultures of caecal contents.

MATERIAL AND METHODS

Six broiler rabbits, Hyplus breed, and six coypus were fed *ad libitum* a commercial granulated feed containing lucerne meal (30%), wheat bran (22.5%), sunflower meal (17%), oats (9%), barley (8%), sugarbeet pulp (6%) and soyabean meal (3%) as the main ingredients. Rabbits and coypus were slaughtered at the age of 3 and 6 months, respectively, between 6:30 and 8:00 a.m. After slaughtering, the stomach, small intestine, caecum and colon were isolated by ligating organs with a string to prevent movement of the digesta. Digestive organs were weighed and their contents squeezed out into beakers. In all samples, the pH was measured and DM concentration determined by drying at 105°C. The caecal contents (10 g) were added to 30 ml of water or Burroughs buffer (BURROUGHS *et al*., 1950) with yeast extract (1 g/l) and urea (0.5 g/l). To stop the fermentation in former samples, $HgCl_2$ was added immediately. The latter samples were incubated with or without a substrate (0.6 g of wheat bran) in 100 ml serum bottles at 39°C for 24 h. The bottles were thoroughly flushed with CO_2 and hermetically closed with rubber stoppers. Sodium sulphide was added to the incubation fluid at 0.5 g/l as a reducing agent. The pH (>7 initially) fell by 0.7-0.9 in the course of the incubation.

Total volatile fatty acids (VFA) were estimated by titration, after steam distillation. Their molar composition was determined on a gas chromatograph using a column of the Chromosorb WAW with 15% SP 1220/1% H_3PO_4 (Supelco) at 140°C. Samples of the headspace gas were taken at the end of the incubation and analysed on the same column. At the same time, the manometric pressure in the incubation vessels was measured. Lactic acid was assayed by the microdiffusion method (CONWAY, 1957). Ammonia was determined colorimetrically with Nessler reagent (after prior separation from interfering compounds by microdiffusion in Conway units) in samples diluted with water and clarified by centrifugation (8000 g for 20 min). Total nitrogen (N) in the caecal contents was determined using the Kjeltec Auto 1030 Analyser (Tecator AB, Sweden).
Metabolic hydrogen balance was calculated according to DEMEYER (1991), except that also the caproate production was taken into account (MAROUNEK *et al*., 2000):

$$2H_{released} = 2A + P + 4B + 3V + 6C$$

$$2H_{accepted} = 4M + 2P + 2B + 4V + 4C$$

$$2H_{recovery} = (2H_{accepted} / 2H_{released}) \cdot 100\%$$

where A, P, B, V, C and M represent molar production of acetate, propionate, butyrate, valerate, caproate and methane, respectively. Such calculation compares the amounts of metabolic hydrogen produced and recovered in reduced end products. The significance of differences was evaluated by the *t*-test.

RESULTS AND DISCUSSION

At the time of slaughter, the average body weight of coypus was 4.13 ± 0.43 kg and that of rabbits 2.87 ± 0.36 kg. The caecum was the heaviest digestive organ in both animal species (Table 1).

Table 1. Weights of digestive organs of six coypus and six rabbits, digesta dry matter concentration and pH.

	Weight (g)	Dry matter (%)	pH
Coypus			
Stomach	85.1 ± 14.3* (2.1%)	17.6 ± 1.8	3.80 ± 0.65*
Small intestine	110.6 ± 13.9* (2.5%)	13.5 ± 1.3	6.98 ± 0.12
Caecum	169.8 ± 17.8 (4.1%)	17.9 ± 0.6*	6.10 ± 0.28
Colon	115.8 ± 20.2* (2.8%)	19.4 ± 1.7*	6.16 ± 0.25
Rabbits			
Stomach	135.3 ± 15.8 (4.7%)	19.0 ± 3.0	2.39 ± 0.31
Small intestine	89.2 ± 15.9 (3.1%)	11.4 ± 2.9	6.96 ± 0.11
Caecum	156.7 ± 18.4 (5.5%)	24.1 ± 1.9	5.81 ± 0.28
Colon	68.6 ± 13.5 (2.4%)	24.6 ± 2.2	6.10 ± 0.25

Means ± SD.
Values in parentheses are percentages of organ weight from animal weight.
*Significantly different from a corresponding value in rabbits (P<0.05).

Table 2. Caecal metabolites in six coypus and six rabbits.

	Coypus	Rabbits
Total VFA[A]	100.1 ± 25.2	104.8 ± 15.8
Acetate[B]	73.9 ± 4.1*	62.8 ± 4.1
Propionate[B]	18.6 ± 2.2*	10.1 ± 4.1
Butyrate[B]	7.4 ± 2.0*	22.7 ± 3.7
Other VFA[B]	0.1 ± 0.1*	4.4 ± 1.3
Lactate[A]	2.6 ± 0.3*	1.1 ± 0.1
Total N[C]	6.78 ± 0.48*	9.09 ± 1.24
Total N[D]	37.9 ± 2.8	37.7 ± 2.6
Ammonia N[A]	23.3 ± 9.2	25.5 ± 8.0

Means ± SD. [A]µmol/g, [B]mol.%, [C]mg/g, [D]mg/g DM
*Significantly different from a corresponding value in rabbits (P<0.002).

Both stomach weight and relative proportion of stomach weight from the body weight were greater in rabbits than in coypus. The gastric pH was significantly higher and dry matter concentration in the hindhut lower in coypus than in rabbits. Total VFA, total N per 1 g of DM and ammonia concentration in the caecal contents of coypus and rabbits were similar (Table 2).

Ammonia N represented 4.81 and 3.93% of the total N in the caecal contents of coypus and rabbits, respectively. Molar percentages of acetate and propionate were significantly higher and percentage of butyrate lower in caecal VFA of coypus than in rabbits. The caecal contents of coypus contained more lactate ($P<0.05$).
Caecal microorganisms of coypus produced significantly more propionate and methane, and less butyrate and valerate than microorganisms of rabbits (Table 3).

Table 3. Production of volatile fatty acids, methane, gas and hydrogen recovery in cultures of caecal contents of coypus and rabbits.

	Substrate[A]	Coypus	Rabbits
Total VFA[B]	+	97.6 ± 17.2	81.2 ± 8.6
Acetate[C]	+	64.1 ± 2.8	60.3 ± 3.3
Propionate[C]	+	21.3 ± 1.9*	9.7 ± 1.0
Butyrate[C]	+	11.1 ± 0.9*	22.3 ± 1.6
Valerate[C]	+	0.7 ± 0.1*	2.6 ± 0.7
Caproate[C]	+	0*	1.6 ± 0.6
Isoacids[C]	+	2.8 ± 0.5	3.5 ± 2.0
Methane[B]	+	17.3 ± 2.7*	8.7 ± 2.4
Gas[D]	+	1.46 ± 0.37	1.64 ± 0.14
2H recovery[E]	+	74.3 ± 6.9*	53.2 ± 7.8
Total VFA[B]	–	50.5 ± 10.6	36.8 ± 8.7
Acetate[C]	–	66.0 ± 2.2*	61.1 ± 1.0
Propionate[C]	–	19.5 ± 1.3*	10.7 ± 1.1
Butyrate[C]	–	9.7 ± 0.8*	18.5 ± 1.1
Valerate[C]	–	1.5 ± 0.7*	3.1 ± 1.1
Caproate[C]	–	0*	1.2 ± 0.4
Isoacids[C]	–	3.3 ± 2.22	5.4 ± 0.8
Methane[B]	–	10.8 ± 1.5*	4.9 ± 1.3
Gas[D]	–	0.51 ± 0.23	0.80 ± 0.19
2H recovery[E]	–	80.7 ± 6.7*	62.4 ± 12.3

Means of 4 cultures ± SD. [A]Wheat bran (0.6 g/40 ml), [B]µmol/g, [C]mol.%, [D]ml/l, [E]%.
*Significantly different from a corresponding value in rabbits ($P<0.05$).

Small amount of caproate was produced in caecal cultures of rabbits but not in those of coypus. Hydrogen recovery in latter cultures was significantly higher. Methane represented 28.7 and 12.8% (v/v) of the headspace gas in substrate-supplied cultures of the coypu and rabbit caecal contents, respectively. In substrate-free cultures the total production of VFA, gas and methane were decreased ($P<0.05$). This was accompanied

with a non-significant increase of 2H recovery and marginal shifts in molar proportions of individual VFA. The butyrate molar percentages, however, were significantly lower in rabbit caecal cultures without the substrate. Methane represented 51.2 and 14.8% of the headspace gas in substrate-free cultures of the coypu and rabbit caecal contents, respectively.

A knowledge on digestive physiology of coypus is poor. Considering the large caecum, one could suppose a great similarity between coypus and rabbits, which are the best-studied caecum fermenters. This assumption was not verified in this study. A noteworthy difference in the digestive morphology of coypus and rabbits was a smaller size of the stomach in former animals, which may be the consequence of a limited extent of the caecotrophy. Low DM concentration in the coypu caecal and colonic contents probably reflects little need for the conservation of water. Significantly lower DM concentration in the coypu caecal contents was observed also in our previous study (MAROUNEK *et al*., 1999).

The caecal fermentation pattern in coypus and rabbits differed. High production of propionate and methane, and, consequently, high 2H recoveries in the coypu caecal cultures indicate that different major hydrogen sinks (methanogenesis *vs* synthesis of acetate from CO_2 and H_2) exist in the caeca of coypus and rabbits. This may be caused by the composition of nutrients leaving the small intestine and/or by different microorganisms colonizing the caeca. One mmol of CH_4 was produced per 5.64 and 9.33 mmols of VFA in the coypu and rabbit caecal cultures with substrate, respectively. Corresponding values in cultures without a substrate were 4.68 and 7.51. The lack of fermentable substrate thus increased the relative importance of caecal methanogenesis, as observed also by PIATTONI *et al.* (1997).

CONCLUSION

Caecal fermentation pattern differed in coypus and rabbits more than other parameters examined in this study or reported by other authors.

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