EFFECT OF LOW-DIGESTIBLE CARBOHYDRATES ON CAECAL AMMONIA CONCENTRATION IN RATS AND TURKEYS

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Summary. The work comments on the results of a series of experiments on caecal ammonia concentration in rats and turkeys fed diets containing different types and doses of low-digestible carbohydrates (LDC). Dietary lactulose and inulin (4-8%) added to a diet for rats effectively reduced the caecal ammonia concentration compared to the cellulose or sucrose control group. A strong adverse effect was observed in the case of 4-5% oligosaccharides from lupin and pea seeds. Dietary 5% xylitol and β-galactosyl-derivatives of sugar alcohols slightly enhanced or had no effect on caecal ammonia concentration in rats. In young turkeys, small doses (0.1-0.4%) of low-digestible carbohydrates were sufficient to decrease ammonia concentration in the caeca, while in older birds that effect was not observed.
Keywords: low-digestible carbohydrates, ammonia, caecal digesta, rat, turkey.

SUNKIAI VIRŠKINAMŲ ANGLIAVANDENIŲ POVEIKIS ŽIURKIŲ IR KALAKUTŲ AKLOSIOS ŽARNOS AMONIAKO KONCENTRACIJAI

Santrauka. Bandymo tikslas – įvertinti poveikį žiurkių ir kalakutų, šertų ir lesintų racionais su skirtingų tipų sunkiai virškinamais angliavandeniais bei skirtingais jų kiekiais, aklosios žarnos amoniako koncentracijai. Į žiurkių pašarus įtraukti maistiniai laktulozė ir inulinas (4–8%) veiksmingai sumažino aklosios žarnos amoniako koncentraciją palyginti su celiuliozės arba sacharozės gavusiomis kontrolinėmis grupėmis. Ryškus priešingas 4–5% poveikis buvo pastebėtas šeriant oligosacharidais iš lubinų ir žirnių. Maistinis 5% ksilitolis ir betagalaktosilis cukraus alkoholių derivatus šiek tiek padidino arba neturėjo jokio poveikio žiurkių aklosios žarnos amoniako koncentracijai. Jauniems kalakutams mažų (0,1–0,4%) sunkiai virškinamų angliavandenių dozių pakako amoniako koncentracijai aklojoje žarnoje sumažinti, tačiau vyresniems paukščiams toks poveikis nenustatytas.
Raktažodžiai: sunkiai virškinami angliavandeniai, amoniakas, akloji žarna, žiurkės, kalakutai.

Introduction. Some studies have reported that supplementation of a diet for laboratory animals as well as for poultry with low-digestible oligosaccharides may bring beneficial changes to the metabolism of the gastrointestinal tract (Monsan and Paul, 1995). Although differing in their chemical characteristics, all the LDC resist digestion (to a different extent) in the small intestine of humans and monogastric animals, and they are potential substrates for the bacteria colonizing the large intestine (Campbell et al., 1997). Many of putrefactive compounds have adverse effects on large intestinal health, e.g. high ammonia concentration may promote tumorigenesis (Lin and Visek, 1991). The fermentation of carbohydrates affects nitrogen metabolism in the large intestine. It has been reported that ammonia concentration in the gut lumen can be reduced by active carbohydrate fermentation, which stimulates the bacterial requirement for nitrogen due to an enhanced growth (Jorgensen et al., 2003). The work comments on the results of a series of own experiments on caecal ammonia concentration in growing rats and turkeys fed diets containing different types and doses of LDC.

Material and Methods. All experiments were conducted on growing Wistar rats and three-day-old BUT-9 turkeys. In studies with rats, the control groups were fed with a casein diet containing adequate amount of cellulose or sucrose, and experimental groups were fed with a diet in which cellulose or sucrose was replaced with LDC preparations. The control diet contained about 13.5% crude protein (casein with DL-methionine), a standard amount of mineral mixture (according to AIN-93G Mineral Mix) and vitamin mixtures (according to AIN-93G Vitamin Mix), 10% soya oil, and maize starch as the rest. In experiments with turkeys, the basal diet was formulated to meet the nutrient requirements for birds (NRC, 1994) and in long-term studies it was changed every four weeks. The LDC preparations were given instead of wheat or maize.

At the termination of the experiments, rats and turkeys were killed according to the recommendation for euthanasia of experimental animals (Close et al., 1997). Samples of fresh caecal digesta were used for immediate analysis of ammonia concentration. In fresh caecal digesta ammonia extracted and trapped in a solution of boric acid in Conway's dishes was determined by direct titration with sulphuric acid (Hofirek and Haas, 2001). The results were analysed using one-way ANOVA and significant differences between groups were determined by the Duncan's multiple range test. Differences were considered significant at $P \leq 0.05$.

Results and Discussion. Caecal ammonia concentration means from studies on rats using cellulose or sucrose negative control *vs.* LDC-supplemented diets are shown in Table 1. In our studies, the LDC preparations were compared with sucrose – a dietary component digested and absorbed in the small intestine,

and with cellulose as an inert polysaccharide having little effect on the caecum in rats (Brunsgaard et al., 1995). The results showed that both the type and dose of LDC strongly influenced caecal ammonia concentration. In studies with dietary lactulose and inulin ammonia concentration in the caecum was observed to decrease beneficially. The experiment with 8%-lactulose-syrup diet (Juśkiewicz and Zduńczyk, 2002) showed 40% reduction in ammonia concentration compared to the cellulose group. In that study, half the rats fed a diet containing 8% of lactulose showed symptoms of diarrhoea and the ammonia concentration in the caecum was enhanced by diarrhoea ailment. The authors pointed out that the occurrence of diarrhoea significantly deteriorated functioning of the caecal ecosystem, which in turn limited potential benefits of diet supplementation with lactulose. The administration of lactulose to humans caused an increase in faecal nitrogen and a reduction of luminal ammonia concentrations (Scheppach et al., 2001). Lactulose has been used worldwide as a drug for the treatment of hepatic encephalopathy and hyperammonaemia. In the other study (Zduńczyk et al., 2004), the administartion of crystalline-lactulose and inulin (4 or 8 % of the diet) was also accompanied by a decrease in the ammonia concentration in the caecal digesta, compared to the cellulose or sucrose control. The effect was the most pronounced with 8% lactulose preparation.

Table 1. **Ammonia concentration in caecal digesta of rats fed diets containing different low-digestible carbohydrates**

Age [weeks]	LDC use rate [%] and feeding period	Negative control [mg/g]	LDC [mg/g]	Difference[#] [%]	Reference
9	syrup-lactulose[1], 8 %, 4 weeks	0.40 [cel]	0.24	-40.00*	Juśkiewicz and Zduńczyk, 2002
9	syrup-lactulose[2], 8 %, 4 weeks	0.40 [cel]	0.31	-22.50	Juśkiewicz and Zduńczyk, 2002
8	crystalline-lactulose, 4%, 4 weeks	0.45 [suc] 0.62 [cel]	0.40 0.40	-11.11 -35.48*	Zduńczyk et al., 2004
8	crystalline-lactulose, 8%, 4 weeks	0.45 [suc] 0.62 [cel]	0.27 0.27	-40.00 -56.45*	Zduńczyk et al., 2004
8	inulin, 4%, 4 weeks	0.45 [suc] 0.62 [cel]	0.34 0.34	-24.24 -45.16*	Zduńczyk et al., 2004
8	inulin, 8%, 4 weeks	0.45 [suc] 0.62 [cel]	0.36 0.36	-20.00 -41.94*	Zduńczyk et al., 2004
8	lupin oligosaccharides, 3.89 %, 2 weeks	0.24 [cel]	0.51	+112.5*	Juśkiewicz et al., 2003a
8	pea oligosaccharides, 4.87 %, 2 weeks	0.24 [cel]	0.46	+91.67*	Juśkiewicz et al., 2003a
8	inulin, 5 %, 4 weeks	0.55 [suc]	0.45	-18.18	Juśkiewicz et al., 2004a
8	xylitol, 5 %, 4 weeks	0.55 [suc]	0.59	+7.27	Juśkiewicz et al., 2004a
8	β-gal-xylitol, 5 %, 4 weeks	0.55 [suc]	0.57	+3.64	Juśkiewicz et al., 2004a
8	β-gal-sorbitol, 5 %, 4 weeks	0.55 [suc]	0.62	+12.73	Juśkiewicz et al., 2004a
8	β-gal-erythritol, 5 %, 4 weeks	0.55 [suc]	0.54	-1.82	Juśkiewicz et al., 2004a

[#]change relative to negative control set at 100 % * P≤0.05
[1]rats without diarrhoea symptoms [2]rats with diarrhoea symptoms

In the study with dietary oligosaccharides from lupin and pea seeds (α-galactosides), an adverse effect on caecal ammonia in rats was observed (Juśkiewicz et al., 2003a). The increase of ammonia concentration can be explained by two factors: (1) oligosaccharide preparations were also an additional source of protein in the diets; and (2) since oligosaccharides inhibit the protein digestibility in the upper part of the gastrointestinal tract (Gdala et al., 1997), a part of dietary protein could escape digestion in the small intestine and was digested by bacteria in the caecum, thus increasing the ammonia concentration. The lack of reduction in the caecal ammonia concentration was also observed in the recent study with dietary xylitol and β-galactosyl-derivatives of sugar alcohols (Juśkiewicz et al., 2004a). Compared to the control rats fed sucrose, the concentration of ammonia in the caecum was found to decrease only in the case of inulin, and to increase in the case of β-galactosyl-sorbitol preparation.

Considering that the use of antibiotics for growth promotion in poultry has been banned in many countries,

the diet supplementation with oligosaccharides could act as a replacement for antibiotics in diets (Spring, 1999). Some studies have shown that even small doses of oligosaccharides (0.1-0.4%) added to a diet were effective in improving health and production of poultry (Savage and Zakrzewska, 1997). Caecal ammonia concentrations of turkeys fed diets supplemented with different oligosaccharides are presented in Table 2. The results suggested that in young birds small amounts (0.1-0.4%) of different LDC, e.g. FOS, inulin, MOS, effectively reduced ammonia concentration in the caeca (Juśkiewicz et al., 2002; 2003). On the other hand, in long-term feeding studies such an effect was not observed in the case of inulin, even at a supplementation dose of 1.0% of a diet (Juśkiewicz et al., 2004). When different levels of MOS were administered for 16 weeks to commercial turkeys, the highest ammonia concentration in the caecal digesta was associated with a low dose (0.1%) of mannan in a diet, and it was reduced to the control level when the highest dose of MOS was used (Zduńczyk et al., 2005).

Table 2. **Ammonia concentration in caecal digesta of turkeys fed diets containing different low-digestible carbohydrates**

Age♣ [weeks]	LDC use rate [%]	Negative control [mg/g]	LDC [mg/g]	Difference# [%]	Reference
4	FOS, 0.4 %	0.66 [wheat]	0.55	-16.67*	Juśkiewicz et al., 2002
4	inulin, 0.4 %	0.66 [wheat]	0.54	-18.18*	Juśkiewicz et al., 2002
4	mannan, 0.1 %	0.66 [wheat]	0.58	-12.12*	Juśkiewicz et al., 2003
4	mannan, 0.2 %	0.66 [wheat]	0.57	-13.64*	Juśkiewicz et al., 2003
4	mannan, 0.4 %	0.66 [wheat]	0.55	-16.67*	Juśkiewicz et al., 2003
16	inulin, 0.1 %	0.91 [corn]	1.11	+21.98	Juśkiewicz et al., 2004
16	inulin, (0.4/0.2 %)[1]	0.91 [corn]	1.19	+30.77*	Juśkiewicz et al., 2004
16	inulin, (1.0/0.4 %)[2]	0.91 [corn]	1.24	+36.26*	Juśkiewicz et al., 2004
16	mannan, 0.1 %	0.91 [corn]	1.18	+29.67*	Zduńczyk et al., in press
16	mannan, (0.4/0.2 %)[1]	0.91 [corn]	1.09	+19.78	Zduńczyk et al., in press
16	mannan, (1.0/0.4 %)[2]	0.91 [corn]	0.98	+7.69	Zduńczyk et al., in press

♣also feeding period #change relative to negative control set at 100 % * P≤0.05

[1]0.4 and 0.2 % in the first and second 8-week period, respectively

[2]1.0 and 0.4 % in the first and second 8-week period, respectively

Conclusions

The effect of dietary LDC on the caecal ammonia concentration observed in these studies on rats and turkeys was strongly determined by the type and amount of dietary carbohydrate preparation as well as the age of animals.

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