Effects of probiotic bacteria on the performance of arctic foxes, pathomorphology and microflora of their alimentary tracts

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ABSTRACT: Studies on the use of *Enterococcus faecium* and *Lactobacillus acidophilus* cultures in arctic fox nutrition were performed on 80 foxes, in the period from weaning to the end of growth and fur development. Diets for the experimental animals were supplemented with 1 g of a probiotic preparation that provided the supply of each bacterial culture in the amount of 1×10^9 CFU (colony forming units) per day. The performance of the animals was estimated on the basis of their body weights, conformation and pelt quality. Histopathological examinations of the liver, kidneys and selected segments of the alimentary tract were also conducted. The results show that a mixture of *Enterococcus faecium* and *Lactobacillus acidophilus* cultures had beneficial effects on the health of artic foxes. The probiotic changed the composition of the alimentary tract microflora, which had a positive influence on the morphology of the alimentary tract mucosa and allowed to reduce morphological damage to the liver and kidneys. The good condition of foxes resulted in higher body weight gains and better parameters of conformation and pelt quality.

Keywords: arctic fox; nutrition; probiotic; growth; pelt; alimentary tract; microflora

Probiotics are preparations containing an amount of strictly defined microorganisms that colonize the alimentary tract, in this way improving the microflora of a given region in the host organism, which has positive effects on health. In the case of fur-bearing animals, such probiotic microorganisms are first of all lactic acid bacteria: *Lactobacillus, Bifidobacterium* and *Enterococcus,* as well as yeast-like fungi *Saccharomyces* (Tauson, 1983, 1984; Jorgensen, 1988a,b; Jorgensen and Pedersen, 1992; Pedersen and Jorgensen, 1992; Balakiriev *et al*., 1994; Lorek *et al*., 2000). The mechanism of their action is multi-directional. They acidify the intestinal contents, inhibiting the growth of pathogenic bacteria, compete with other microorganisms for nutrients and receptors, or adhere to epithelial cells, producing antibacterial compounds, and affect the immune system. The research results show that *Enterococcus faecium* is most suitable for fur-bearing animals (Jorgensen, 1988b; Jorgensen and Pedersen, 1992; Pedersen and Jorgensen, 1992; Simon *et al*., 2001). However, professional literature does not provide a full description of the alimentary tract microflora in carnivorous fur-bearing animals. Williams *et al*. (1998) stressed that the alimentary tract microflora of minks was different, compared to other animal species. According to these authors, this is connected with their diet, including feed of animal origin, and a considerable rate of its transportation in the alimentary tract. They believe that the microflora composition changes with age and depends on the feed ration. It is important to maintain the right proportions between particular groups of microorganisms typical not only of particular species but also of alimentary tract segments.

MATERIAL AND METHODS

The experimental factors were lyophilised and microcapsuled bacterial cultures of *Enterococcus faecium* and *Lactobacillus acidophilus* contained in the preparation ALL-LAC (manufacturer – Alltech Biotechnology Center Inc., USA). The experiment was performed on 80 arctic foxes, aged between 8 and 20 weeks, i.e. in the period from weaning to the end of growth and fur development. They were divided into two equal groups, taking into account their sex and origin, and placed in conventional production cages (four animals in each). The animals were kept under good sanitary conditions in accordance with welfare standards. The experimental and control foxes were fed *ad libitum,* and the nutritive value of diets fully satisfied their requirements. The rations contained the following components: raw meat offal (20–30%), preserved meat offal (0–5%), hard poultry offal (30–35%), ground barley (18–20%), wheat bran (2–3%), steamed potatoes (0–12%), green forage and vegetables (11%). The proportions of particular components changed in dependence on the feeding period. The foxes from the control group (I) were fed the same diets as the other animals on the farm. The foxes from the experimental

Table 1. Performance indices in arctic foxes

Specification	Statistical measures	Group I	Group II
Body weight			
Initial body weight (kg)	n	40	40
	$\bar{x}$	1.92	1.94
	v	10.27	12.22
Final body weight (kg)	$\bar{x}$	6.94	7.21
	v	10.75	11.77
Results of conformation evaluation			
Trunk length (cm)	$\bar{x}$	63.75	64.45
	v	4.50	3.42
Animal size and constitution (points)	$\bar{x}$	4.75	5.30
	v	32.60	21.13
Colour type (points)	$\bar{x}$	2.97	3.00
	v	5.31	0.00
Colour purity (points)	$\bar{x}$	2.55^{B}	2.87^{A}
	v	19.76	11.65
Fur quality (points)	$\bar{x}$	5.97^{B}	6.47^{A}
	v	17.57	16.02
Total score (points)	$\bar{x}$	16.24^{B}	17.64^{A}
	v	13.03	9.89
Pelt evaluation and classification			
Size (cm)	$\bar{x}$	107	109
	v	6.76	4.71
Fur category	$\bar{x}$	2.03	1.76
	v	36.57	44.26

$^{a,\,b}P < 0.05$; $^{A,\,B}P < 0.01$; n = number of animals; $\bar{x}$ = mean; v = coefficient of variation (%)

group (II) were given diets supplemented with 1 g of a probiotic preparation, which provided the supply of each bacterial culture in the amount of 1×10^9 CFU (colony forming units) per day. The body weights of the animals were determined at the beginning and at the end of the experimental period by individual weighing. Their conformation was evaluated at the end of fur development, according to the Standards of Conformation Evaluation in Arctic Foxes (1998). Pelt evaluation and classification by size and fur category (Polish Standard, 1984) were made after slaughter and pelt processing. Histopathological examinations were conducted on three males and three females from each group. Sections of the liver, kidneys, stomach, duodenum, jejunum and colon were fixed in 10% formalin and paraffin embedded. The microtome sections were haematoxylin and eosin (HE) and PAS stained (according to McManus). Samples of the stomach, jejunum and colon were taken for examinations of the alimentary tract microflora. 2 g of tissue from each sample was homogenized to make a 20% suspension in sterile physiologic saline. Each sample was diluted 10 times in sterile broth and inoculated in two replications. The Mueller-Hinton agar and agar with glucose were used to determine the total count of aerobic bacteria. The Edwards medium was used for the determination of the enterococcus count, and blood agar – for the isolation of *Escherichia coli*. The Wrzosek broth – to isolate anaerobic bacteria. The cultures were incubated in a thermostat at 37°C for 18–72 hours. After incubation the number of colonies on the media was determined, together with the geometric mean bacterial count per g of the examined tissue. The numerical data were analysed statistically by an analysis of variance for one-factor orthogonal designs (Statistica 6.0 Software PL).

RESULTS AND DISCUSSION

The initial body weights were similar in both groups (Table 1). The final body weights of the experimental animals were by 0.27 kg higher, but this difference was statistically insignificant. In her studies on minks and foxes Tauson (1983, 1984) observed higher body weight gains in groups given probiotics. Balakiriev *et al*. (1994) also reported positive effects of probiotic cultures on body weight gains in minks and foxes. Dahle and Holstad (1988) did not note any influence of lyophilised *Enterococcus faecium* cultures added to pellets on body weights of arctic foxes. The results of conformation evaluation (Table 1) confirm that the experimental animals received more points for size and body constitution. They were also longer (measurements taken with a tape) than the control ones. This indicates that higher body weights of the experimental foxes, although not confirmed statistically, were correlated with their size and trunk length. The colour type did not show any statistical differences. The fur colour purity was higher in the experimental group. The colour type is determined genetically, whereas the fur colour purity depends on environmental factors, including diet. The fur quality in group II was statistically highly significantly better than in group I. The total score for conformation was also statistically highly significantly higher in group II. It was connected with better fur quality and bigger size. The pelt length was higher in group II, which was correlated with the final body weight and conformation. As regards fur quality, the pelts of the experimental foxes (group II) were close to the first category. This result was consistent with those obtained in the case of conformation evaluation. This is natural, as the conformation evaluation is based on fur parameters that also decide about pelt quality. Jorgensen (1988a) reported better pelt quality in minks given *Enterococcus faecium* cultures. Balakiriev *et al*. (1994) observed positive effects of Bifidubakterin on pelt quality in arctic foxes, common fixes and minks. Lorek *et al*. (2000) noted no influence of probiotic bacteria and yeast on the size and quality of mink pelts. Histopathological examinations of the liver (Table 2) showed congestion, parenchymatous and vacuolar degeneration of hepatic cells. These changes were observed in a larger number of animals in the control group. A similar direction of changes was also found in the kidneys. The investigations conducted by Lorek *et al*. (2000) indicated that the internal organs of minks fed diets with a probiotic preparation were better supplied with blood, and their liver showed lower tendencies towards fat deposition. In the animals from both groups the gastric mucosa was congested. Other changes were more intensive in group I. Acute catarrhal inflammation of the mucosa was observed in the duodenum. Some of the control animals showed excessive exfoliation of epithelial cells and deformed villi. In the experimental animals they were usually regular and longer. The inflammation was less intense in the further segments of the small intestine, especially in the experimental group. Also

Table 2. Results of morphological and histopathological examination of selected internal organs and selected segments of the alimentary tract

Organs or segments of the alimentary tract	Changes	Group	
		I	II
Liver	congestion, haemostasis	5	1
	parenchymatous degeneration	6	3
	vacuolar degeneration	6	1
Kidneys	congestion, haemostasis	6	3
	parenchymatous degeneration	6	4
	vacuolar degeneration	6	1
Stomach	congestion of the mucosa	6	5
	congestion of the submucosa	6	0
	catarrhal inflammation of the mucosa	1	0
	excessive exfoliation of epithelial cells	4	1
Duodenum	congestion of the mucosa	6	1
	congestion of the submucosa	5	1
	catarrhal inflammation of the mucosa	6	1
	deformed villi	6	3
	excessive exfoliation of epithelial cells	6	3
Jejunum	congestion of the mucosa	4	1
	congestion of the submucosa	4	1
	catarrhal inflammation of the mucosa	3	1
	deformed villi	6	1
	excessive exfoliation of epithelial cells	4	2
Colon	congestion of the mucosa	0	0
	congestion of the submucosa	6	4
	catarrhal inflammation of the mucosa	2	0

the villi in the jejunum and ileum were more regular and longer in this group. The colon submucosa was congested in all animals from the control group. In group II its congestion was noted in four cases only. The inflammatory process in the duodenum and jejunum could be the reason for higher intensity of morphological changes in the control foxes, in the form of parenchymatous and vacuolar degeneration of hepatic cells and renal tubule epithelium. The results obtained by Lorek *et al*. (2000) show that there were no morphological changes in the alimentary tracts of minks fed diets supplemented with a probiotic preparation, except for increased exfoliation of intestinal mucosa epithelium. In the group receiving probiotic supplementation the count of potentially pathogenic *Escherichia coli* rods was considerably lower (Table 3), whereas that of *Enterococcus* – considered a natural antagonist of *Escherichia coli* – was higher (Jorgensen, 1988b; Simon *et al*., 2001). Such a microbiological state of the alimentary tract may affect positively the general health condition of foxes. The experiments performed on dogs (whose alimentary tract resembles that of foxes) indicate that diet supplementation with *Enterococcus faecium* cultures causes an increase in the count of enterococci in the ileum contents and faeces (Molitor, 1996; Zentek *et al*., 1998).

Table 3. Mean values of the algorithms of the microorganism count in selected segments of the alimentary tract

Group	Segment of the alimentary tract	Total aerobe count	*Coli* bacteria	*Enterococcus*	Spore-forming anaerobes
I	stomach	3.3	2.3	1.7	–
	jejunum	4.2	3.6	2.1	–
	colon	6.6	5.7	3.5	2.1
II	stomach	2.9	1.9	2.9	–
	jejunum	4.3	2.5	4.3	–
	colon	6.5	4.3	5.9	2.3

CONCLUSIONS

The obtained results show that a mixture of *Enterococcus faecium* and *Lactobacillus acidophilus* cultures had beneficial effects on the health of arctic foxes. The probiotic changed the composition of the alimentary tract microflora, which had a positive influence on the morphology of the alimentary tract mucosa and allowed to reduce morphological damage to the liver and kidneys. Good condition of the foxes resulted in higher body weight gains and better parameters of conformation and pelt quality.

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ABSTRAKT

Vliv probiotických bakterií na užitkovost polárních lišek, patomorfologii a mikroflóru jejich trávicího ústrojí

V období od odstavu do ukončení růstu a vytvoření kožešiny byl sledován účinek kultur *Enterococcus faecium* a *Lactobacillus acidophilus* ve výživě arktických lišek (n = 80). Do krmných směsí pro pokusná zvířata byl přidán 1 g probiotického preparátu, který zajišťoval dávku bakteriální kultury v množství 1 × 10^9 CFU (kolonu tvořících jednotek) na den. Užitkovost zvířat byla hodnocena na základě jejich tělesné hmotnosti, exteriéru a kvality kožešiny. Rovněž bylo provedeno vyšetření jater, ledvin a vybraných částí trávicího ústrojí. Výsledky naznačují, že směs kultur *Enterococcus faecium* a *Lactobacillus acidophilus* měla blahodárné účinky na zdravotní stav polárních lišek. Probioticum změnilo složení mikroflóry trávicího ústrojí, což mělo kladný vliv na morfologii mukózy trávicího ústrojí, a přispělo ke snížení morfologického poškození jater a ledvin. Dobrá tělesná kondice lišek vedla k vyšším přírůstkům tělesné hmotnosti a lepším ukazatelům exteriéru i k vyšší kvalitě kožešiny.

Klíčová slova: polární liška; výživa; probiotikum; růst; kožešina; trávicí ústrojí; mikroflóra

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