Pet food and feed applications of inulin, oligofructose and other oligosaccharides

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Prebiotics may be considered as functional food ingredients. They are attracting considerable interest from pet owners, pet food manufacturers, livestock producers and feed manufacturers. The most common forms of prebiotics are nondigestible oligosaccharides (NDO), including inulin, oligofructose mannanoligosaccharides, gluco-oligosaccharides, and galacto-oligosaccharides. These NDO are nondigestible by enzymes present in the mammalian small intestine, but are fermented by bacteria present in the hindgut of nonruminants. Inulin and oligofructose are present in measurable quantities in feed ingredients like wheat, wheat by-products, barley, and peanut hulls. Consumption of prebiotic oligosaccharides elicits several purported health benefits. In companion animals, prebiotics have been shown to improve gut microbial ecology and enhance stool quality. In production livestock and poultry, prebiotics are employed to control pathogenic bacteria, reduce faecal odour, and enhance growth performance. Research to date indicates positive effects of prebiotics on health status and performance of companion animals, livestock, and poultry.

Prebiotics: Fructans: Animal nutrition

Introduction

There currently exists much interest in the use of prebiotics for companion animals, livestock, and poultry as modulators of colonic bacterial populations and fermentation end-products. Prebiotics are nondigestible food ingredients that positively affect the host by selectively stimulating the activity of a limited number of beneficial colonic bacteria, resulting in improved host health (Gibson & Roberfroid, 1995). Inulin and oligofructose are perhaps the most well studied prebiotics (for a description of these products see Roberfroid, 2002).

Concentrations of inulin and oligofructose in pet food and feed ingredients

Hussein *et al.* (1998) selected twenty-five common ingredients and analysed them for oligofructose concentrations (Table 1). In this study, the concentrations of three major subcomponents of oligofructose (1-kestotriose, 1,1-kestotetraose, and 1,1,1 kestopentaose) were assayed via anion exchange HPLC. No oligofructose was detected in corn, corn distiller's solubles, hominy, milo, brown rice, white rice, brewer's rice, rice hulls, seaweed, or soybean meal. On a dry matter basis, wheat co-products (bran, germ,

and middlings) contained the highest concentrations of total oligofructose, followed by peanut hulls, alfalfa meal, barley, and wheat. The remaining ingredients contained very low concentrations (< 0.4 mg/g) of oligofructose. While this database provides information on a number of commonly used pet food ingredients, additional analysis of components of oligofructose with longer degree of polymerization (DP) and inulin concentrations would be desirable.

Van Loo et al. (1995) reported the concentrations of inulin and oligofructose in common dietary ingredients (Table 1). Their analyses quantified glucose and fructose released by enzymatic hydrolysis of the food or plant material and assayed oligofructose (DP up to ten) and inulin contents with a DP of two to sixty (Quemener et al. 1994). Values are reported on an 'as-is' basis and indicate the range of concentrations determined due to variation in sources of each food or plant material. Garlic contained the highest concentration of oligofructose. Wheat and dried onion contained similar amounts of oligofructose, while rye flour and barley contained the lowest concentrations. The authors concluded that inulin and oligofructose are present in significant amounts in a wide variety of common foods and food ingredients.

Abbreviations: CE, competitive exclusion; MOS, mannanoligosaccharides; NDO, nondigestible oligosaccharides; TOS, transgalacto-oligosaccharides. Note: For the definition of the terms inulin and oligofructose please refer to the introductory paper (p. S139) and its footnote.

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ltem	Oligofructose content* mg/g (dry matter basis)	Inulin and oligofructose content† mg/g (as-is basis)
Alfalfa meal	2.24	
Barley	1.92	0.05-0.10
Beet pulp	0.05	
Canola meal	0.04	
Corn aluten feed	0.09	
Corn gluten meal	0.34	
Garlic		0.98-1.60
Oats	0.36	
Oat groats	0.12	
Onion. dried		0.11-0.75
Peanut hulls	2.40	
Rice bran	0.14	
Rve flour		0.05-0.10
Sovbean hulls	0.12	
Wheat	1.36	0.10-0.40
Wheat bran	4.00	
Wheat germ	4.68	
Wheat middlings	5.07	

 Table 1. Inulin/oligofructose content of selected feeds, pet foods and food ingredients

* Determined by Hussein *et al.* (1998) as sum of 1-kestotriose + 1,1-kestotetraose + 1,1,1-kestopentaose.

† Determined by Van Loo et al. (1995) as sum of inulin and oligofructose with DP of 2 to 60.

Applications of inulin and oligofructose in animal nutrition

Perhaps the best-known nutritional effect of inulin and oligofructose is their ability to modify the composition of the intestinal microflora (e.g. increased numbers of bifidobacteria) and their metabolic activity in the large intestine (Roberfroid et al. 1998; Van Loo et al. 1999). In general, most gut bacteria can be divided into groups that exert detrimental effects (staphylococci, clostridia, and veillonella) or those that benefit the host (bifidobacteria, lactobacilli, and eubacteria). Detrimental influences include diarrhoea, infection, and digesta putrefaction, as well as the absorption and metabolism of mutagenic and carcinogenic chemicals (Rowland et al. 1985). Conversely, beneficial bacteria bestow positive effects on the host and may inhibit the growth of harmful bacteria, including E. coli, C. perfringens, salmonellae, listeria, campylobacter, and shigellas (Gibson & Wang, 1994; Araya-Kojima et al. 1995). Homma (1988) described bifidobacteria as a resistance factor in humans based on defence against pathogens, infections and reduction of serum cholesterol. Other beneficial effects attributed to bacteria include stimulation of immune function, increased mineral absorption, and synthesis of vitamins (Gibson & Roberfroid, 1995). In addition to their effects on gastrointestinal characteristics and systemic metabolism, inulin and oligofructose have been postulated to enhance performance responses of poultry and rabbits (Ammerman et al. 1988, 1989; Morisse et al. 1993). Finally, there are some microbial species that exert both negative and positive effects (e.g. streptococci, E. coli, and bacteroides) and, therefore, are considered neutral (Gibson & Roberfroid, 1995).

Companion animal studies

Flickinger *et al.* (unpublished data) supplemented adult female hounds with 0, 1, 2, or 3 g oligofructose (from

sucrose)/d in gelatin capsules. The basal diet consisted of 372 g/kg chicken protein, 196 g/kg corn, 195 g/kg brewer's rice, 130 g/kg oil, 40 g/kg beet pulp, 30 g/kg liquid digest, 10 g/kg dried brewer's yeast, and 27 g/kg vitamin-mineral premix. Compared to the control, supplemental oligofructose (from sucrose) (3 g/d) tended (P < 0.10) to decrease faecal concentrations of *Clostridium perfringens* (10.04 v. 9.67 \log_{10} CFU/g) and increased (P<0.05) total aerobes $(8.52 v. 9.32 \log_{10} \text{ CFU/g})$. There were no differences among treatments in total anaerobes, bifidobacteria or lactobacilli. However, results from a second study by the same group differed. Sixteen adult male beagles were fed a corn-based diet with or without 3, 6 or 9 g/kg supplemental oligofructose (from sucrose) for 18 days. Dogs fed the highest level of oligofructose tended (P < 0.10) to have higher concentrations of bifidobacteria (9.80 v. 9.40 \log_{10} CFU/g) in their faeces when compared with the control group. Differences in the results of these two studies may be due to variability in basal diet formulations (meat- v. corn- based diets) and in methods of administering oligofructose (capsule v. incorporation into an extruded diet). Also, the diet used in the second study contained some wheat grain that contained fructans.

In a recent study from our laboratory, Swanson *et al.* (unpublished data) fed ileally cannulated dogs a meatbased premium diet supplemented either with 1 g/d oligofructose (from sucrose), 1 g/d mannanoligosaccharides (MOS), 1 g/d oligofructose (from sucrose) + 1 g MOS, or 1 g/d sucrose (control) administered via gelatin capsules. Dogs were adapted to their respective diets for 10 days, followed by a 4-day collection of ileal effluent and faeces in a 4×4 Latin square design. Although neither oligofructose nor MOS altered faecal output, moisture content, or score, MOS decreased (P < 0.05) faecal total aerobes and tended (P = 0.13) to increase faecal concentrations of lactobacilli. Supplemental MOS also increased (P < 0.05) serum lymphocyte concentrations and tended (P = 0.14) to increase serum IgA concentrations. Supplemental oligofructose tended to decrease concentrations of three chemical indicators of faecal odour, tryptamine (P=0.11), tyramine (P=0.15), and indole (P<0.10). From these data, it appears that MOS may favourably alter the composition of the colonic bacteria and elicit a systemic immune response, while oligofructose may reduce faecal odour. However, when combined, oligofructose and MOS did not act synergistically.

Poultry, swine, and rabbit studies

Feeding fructans may be a practical strategy for controlling pathogenic bacteria in chickens. Fukata et al. (1999) fed 1day-old chicks an antibiotic-free diet supplemented either with probiotic bacteria (competitive exclusion; CE), 1g/ kg dietary oligofructose (from sucrose), or probiotics + 1 g/kg oligofructose for 7 days. At 1 day after oral inoculation with 10⁸ CFU of Salmonella enteritidis, only chicks fed CE had fewer (P < 0.05) S. enteritidis (\log_{10} CFU/g) organisms recovered in caecal digesta (2.35 v. 4.57, 4.05, and 2.76 for CE v. control, oligofructose, and CE + oligofructose, respectively). In a second experiment, 1-day-old chicks were administered the same treatments, but were adapted to diets for 21 days prior to inoculation with S. enteritidis. Chicks receiving oligofructose or CE + oligofructose exhibited lower ($P \le 0.05$) caecal concentrations of S. enteritidis at 1 day after inoculation (2.45 and 1.76 v. 4.31 and 4.04 for oligofructose, CE + oligofructose, control, and CE, respectively), suggesting that addition of low levels of oligofructose to the diet of chicks receiving a probiotic may reduce Salmonella colonisation.

Two sources of dietary fructan were evaluated in a study by Chambers *et al.* (1997). Chicks were fed diets supplemented with either no carbohydrates (control), 80 g/kg Jerusalem artichoke flour providing 50 g/kg inulin (JAF), or 50 g/kg refined chicory inulin. Chicks were exposed to *Salmonella* by being reared with seeder chicks gavaged with 10^7 CFU of naladixic acid-resistant *S. typhimurium*. At 6 weeks of age, the average *Salmonella* score of JAFfed chicks was higher (P < 0.05) than that of control-fed chicks, while the score of chicory inulin-fed chicks was lower (P < 0.05) than for other groups. Further research needs to be done to elucidate the effects of different sources of inulin and/or oligofructose on *Salmonella* colonisation of broiler caeca.

In addition to their effects on gastrointestinal characteristics, inulin and oligofructose have been postulated to enhance performance responses of livestock. Houdijk et al. (1998) investigated the effects of oligofructose and transgalacto-oligosaccharides (TOS) in growing pigs. Nine-week-old pigs were fed diets supplemented with either 7.5 or 15 g/kg oligofructose or 10 or 20 g/kg TOS for 6 weeks. The basal diet contained no additional copper, antibiotics, or probiotics. In the first week, supplemental oligosaccharides resulted in decreased (P < 0.05) daily weight gain (control, 953 g; 7.5 g/kg oligofructose, 860 g; 15 g/kg oligofructose, 750 g; 10 g/kg TOS, 765 g; and 20 g/kg TOS, 770 g) and feed conversion efficiency (feed/gain; control, 1.16; 7.5 g/kg oligofructose, 1.23; 15 g/kg oligofructose, 1.35; 10 g/kg TOS, 1.40; and 20 g/kg TOS, 1.24). Dry matter intake was numerically (P=0.10) reduced in oligofructose and TOS treatment groups (control, 1104 g; 7.5 g/kg oligofructose, 1061 g; 15 g/kg oligofructose, 982 g; 10 g/kg TOS, 1036 g; and 20 g/kg TOS, 957 g). During weeks 2 and 3, a similar negative trend occurred, but the differences were not significant. In contrast, during weeks 4-6, oligofructose and TOS supplementation resulted in numerically (P=0.08) increased daily weight gains (control, 861 g; 7.5 g/kg oligofructose, 1056 g; 15 g/kg oligofructose, 981 g; 10 g/kg TOS, 964 g; and 20 g/kg TOS, 1032 g), numerically (P>0.05) greater feed consumption (control, 1655 g; 7.5 g/kg oligofructose, 1852 g; 15 g/kg oligofructose, 1756 g; 10 g/kg TOS, 1850 g; and 20 g/kg TOS, 1830 g) and numerically (P=0.08) enhanced feed conversion efficiency (feed/gain; control, 1.91; 7.5 g/kg oligofructose, 1.73; 15 g/kg oligofructose, 1.83; 10 g/kg TOS, 1.93; and 20 g/kg TOS, 1.78). Differences between the first and last 3-week periods of this study may indicate that young pigs require an adaptation period to dietary oligosaccharides; initially, lower performance can be offset with compensatory growth. The initial depression in feed intake may coincide with fluctuations in colonic microflora ecology, suggesting the role of a non-specific immune response in the observed anorexia.

Other investigators have not found similar effects in swine. Farnworth *et al.* (1992) reported that 15 g/kg oligofructose (from sucrose) or JAF in weanling pig diets did not significantly affect daily feed intake, weight gain, or feed efficiency. However, the authors speculated that the lack of an effect was due to too low a concentration of dietary oligofructose. Similarly, Olsen & Maribo (1999) reported that 16.5 g/kg dietary inulin fed to weanling piglets did not result in a significant difference in average daily weight gain or feed efficiency. The lack of response to supplemental inulin may have been caused by a high fructan content of the basal diet due to wheat and barley inclusion.

In rabbits experimentally infected with *E. coli* O103 and fed either 0 or 2.5 g/kg dietary oligofructose, fewer (P < 0.05) rabbits exhibited clinical signs of enteritis (diarrhoea) in the oligofructose group (14.8 %) as compared to the control group (46.4 %) (Morisse *et al.* 1993). However, mortality rate was not significantly different amongst the treatments (17.9 and 22.2 % for control and oligofructose treatments, respectively). Among surviving animals, oligofructose-fed rabbits tended (P > 0.05) to have heavier body weights (2454 v. 2359 g) and had numerically (P > 0.05) higher average daily weight gains (33.5 v. 32.6 g/d) as compared to the control. While oligofructose supplementation reduced morbidity, it did not significantly improve mortality or growth performance of rabbits.

Future research directions

Many issues remain unresolved concerning prebiotic oligosaccharides, including the establishment of accurate relationships among the composition of the colonic microflora, gastrointestinal tract health and clinical or performance outcomes observed in the animal. Essential to the determination of these relationships is an in-depth understanding of the mechanisms that provide the basis for any observed effect. Another unknown is whether fructans can be used interchangeably. Optimal inulin and/or oligofructose inclusion levels in diets have yet to be established for most animal species. It is unknown whether combinations of different oligosaccharides can elicit diverse beneficial effects, exert a synergistic effect, or perhaps a negative effect. Besides blending prebiotics, synbiotic therapy (coupling probiotic bacteria with prebiotic substrates) perhaps could have a greater impact on the intestinal microflora by nourishing indigenous beneficial bacteria and directly increasing numbers of favourable microbes.

Summary

A relatively small amount of research exists concerning supplementation of fructans and other oligosaccharides in the diets of companion animals, livestock, and poultry. Studies to date indicate a generally positive effect of fructans on colonic microbial ecology, host health, and growth performance. However, more research remains to be done to determine the appropriate role of these oligosaccharides in animal nutrition.

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