PJN
ISSN 1680-5194

PAKISTAN JOURNAL OF NUTRITION

ANSInet
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Pakistan Journal of Nutrition 8 (11): 1756-1759, 2009
ISSN 1680-5194

Effect of Xylooligosaccharide Enriched Yogurt on Serum Profile in Albino Rats

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Abstract: The effect of Xylooligosaccharide (XO) enriched yogurt in addition to basal diet on serum biochemical profile of albino rats assigned to 5 dietary treatments for a period of 21 days were studied. In all groups of rats (n = 5) serum glucose, cholesterol and triglycerides were observed. The weight gain in rats receiving yogurt was higher than those in control group. XO enriched yogurt showed significant improvement in minerals absorption. There was no significant change in serum cholesterol level of rats fed on yogurt with or without XO enrichment but a fair reduction in serum glucose content was noted. XO as a functional ingredient can be incorporated in formulating dairy products with improved health benefits like lowering glucose levels.

Key words: Xylooligosaccharide, yogurt, serum profile, rats

INTRODUCTION

Non Digestible Oligosaccharides (NDO) like Xylooligosaccharides (XOs) have a number of health benefits associated with them like improving gastrointestinal health, preventing of certain diseases such as dental caries, colon cancer and stimulating mineral absorption. Most of the NDOs including XOs are known to have prebiotic characteristics. A prebiotic is defined as a substrate or food ingredient that is non-digestible for the host but is fermented selectively by some of the intestinal microflora (Scholz-Ahrens *et al*., 2001). In this way, it enhances the growth and activity of bacteria with beneficial effects for the host's health. Many of the scientific substantiation for the functional properties of NDOs is supported by animal experiments in which NDOs increased the availability of calcium, magnesium, zinc and iron. This stimulatory effect of some NDOs is assumed to be primarily because of their prebiotic character.

Xylooligosaccharides (XOs) are functional oligosaccharide composed of 2-10 xylose molecules linked together by β (1-4) linkage and are used as food ingredients owing to their technological properties and health effects. XOs are stable over a wide range of pH and temperature enabling them to be used in acidic juices and dairy products like yogurt (Vázquez *et al*., 2000). The main property of the physiological functions results from the indigestibility of XOs, which leads to fermentation in the large intestine, consequently increase *Bifidobacteria* and Short-chain Fatty Acid (SCFA) production. This property results in many types of physiological functions, among which the promotion of mineral absorption, including an increase of bone density and relief of anemia is of particular interest (Hirayama, 2002).

The XOs behave as dietary fibre and since the dietary fibre has the properties mandatory for its consideration as an important ingredient in the preparation of functional foods, due to its beneficial effects such as increasing the volume of faecal bulk, decreasing the time of intestinal transit, cholesterol and glycaemia levels, trapping substances that can be dangerous for the human organism, stimulating the growth of the intestinal flora, etc.

It has also been reported that XOs exhibit some other advantageous consequences on health. Xylooligosaccharides were given in experimental diet to diabetic rats and certain metabolic parameters like blood glucose, serum and liver lipids were examined. The experiment showed that XOs can improve growth retardation, hyperphagia, polydipsia and elevated levels of serum glucose. This is also stated that lowered liver triglycerides and reduction in desaturation index fatty acid composition of liver phosphatidylcholine are outcome of XOs diet. Concluding from above stated observation, it can be recommended that XOs have got properties to be used as sweetener for improving diabetic conditions (Imaizumi *et al*., 1991).

Considering the above stated findings, the present study was conducted to determine the effect of XOs enriched yogurt on the serum biochemical profile in male Albino rats. Since the other XOs are already known to have positive impact on serum biochemical profile, synergistic effect of XOs enriched yogurt was investigated to verify the hypothesis whether synbiotic products have the same impact.

MATERIALS AND METHODS

Study design, animals and diets: Male Albino rats weighing 150±10 g were purchased from the National Institute of Health, Islamabad, Pakistan. The rats were

individually housed in stainless steel wire bottom cages in an environmentally controlled room at 25±2°C and 60±5% relative humidity with 12 h light-dark cycle. The rats were fed on the freshly prepared basal diets for a week and then randomly assigned to 5 dietary treatments for 21 days. Composition of experimental diet is given in Table 1. The detail of the groups on five dietary treatments is given below:

Table 1: Ingredient composition of dietary treatments fed to rats

Ingredients (g/100 g)	Basal diet	XOs enriched diet
Casein	20	20
Mineral mix[1]	3.4	3.4
Vitamin mix[2]	2	2
Sunflower Oil	7	7
Sucrose	7	7
Cellulose	10	5
Corn starch	50	45
Xylooligosaccharide	-	10
Choline bitartrate	0.25	0.25
L-Cystine	0.35	0.35

[1]Mineral (mg/kg): Ca, 5000; P, 1561; K, 3600; Na, 1019; Cl, 1571; S, 300; I, 0.2; Fe, 35; Mg, 507; Zn, 30; Cu, 6; Mn, 10; Mo, 0.15; Se, 0.15; Cr, 1; Si, 5; F, 1; Ni, 0.5; B, 0.5; Li, 0.1 and V, 0.1
[2]Vitamin (mg/kg): thiamin, 5; riboflavin, 6; pyridoxine, 6; nicotinic acid, 30; pantothenate, 15; folic acid, 2; phylloquinone, 0.75; biotin, 0.2; cyanocobalamin, 0.025; all-*trans*-retinyl palmitate (500,000 IU/g), 8; all-*rac*-atocopheryl acetate (500 IU/g), 150 and cholecalciferol (400,000 IU/g), 2.5

- Control group (X_0): Rats fed on basal diet.
- G_1: Rats fed on diet replacing 10% carbohydrate from basal diet with XOs.
- G_2: Rats fed on plain yogurt in addition to basal diet.
- G_3: Rats fed on XOs enriched yogurt (incorporating 3.5% XOs).
- G_4: Rats fed on XOs enriched yogurt (incorporating 4.5% XOs).

XOs were produced in another experiment from corncobs through alkali and enzymatic hydrolysis. Experimental yogurt were prepared according to method described by Mumtaz *et al.* (2008).

All rats were given free access to experimental diets and deionized water. The diet consumed by each rat was determined by deducting the leftover and spilled diet from the total amount supplied per day. Net feed intake of individual rat was measured on daily basis by excluding leftover and collecting spilled diet during the entire period to observe the impact of individual experimental diet. Deionized water was provided with the help of graduated drinking bottles. Gain in body weight of individual rats in each group was determined on weekly basis throughout the experimental period to find out the effect of individual diet on body weight.

Sampling procedures: At the end of the experiment after 21 days, the blood samples were collected from the cecal vein of overnight fasted rats. The serum was separated by centrifugation at 3000 rpm for 15 min after allowing the blood to stand for at least 30 min at room temperature as explained by Uchida *et al.* (2001). After blood sampling, cecum with contents were also removed and weighed. Cecal contents were divided into two halves; one was kept for pH measurement and other one was frozen at -20°C.

- Glucose concentration of individual rats in each group was calculated by GOD-PAP method to determine change in serum glucose level by different diets (Thomas and Labor, 1992).
- Cholesterol in the collected serum of individual rats of all groups was measured by liquid cholesterol CHOD-PAP method to find out the effect of individual diet on the cholesterol level of respective groups Stockbridge *et al.* (1989).
- Triglycerides in the collected serum of individual rats were measured using liquid triglycerides GPO - PAP method as described by Annoni *et al.* (1981).

Statistical analysis: The data obtained was analyzed using Minitab version 14.1. All the experimental groups were compared using analysis of variance (ANOVA). Values in tables and graphs are expressed as mean± SEM. A P value of less than 0.05 was considered to be significant.

RESULTS AND DISCUSSION

The diet intake ranged between 16-20 g/day and with little fluctuation did not differ among all treatment groups. The diet replacing 10% carbohydrate with XOs in basal diet and including plain yogurt in basal diet significantly improved the body weight, but the weight gain was highly significant in rat groups fed on XOs enriched yogurt (Table 2). These results agree with the findings of Imaizumi *et al.* (1991) who found that XO diet caused body weight gain in diabetic rats. The other groups were fed on diet including yogurt (plain and XOs enriched), and the rich nutritional status of the yogurt can be proposed as reason for weight gain in these groups. Moreover, experimental diets had a profound effect on the total weight, wall weight and contents weight of cecum (Table 2). The total weight was the highest in rats fed on XOs diet, which is in agreement of study executed by Campbell *et al.* (1997). The other dietary treatments including plain yogurt and XOs enriched yogurt also caused more cecal weight than that of control group. The cecum weight was double in rats fed on XOs enriched yogurt. This is possibly because of XOs diet leading towards production of Short Chain Fatty Acids (SCFA) in the large intestine and these SCFA serve as energy source to increase cell density along with regularized cell proliferation. Yogurt is a low pH food and SCFA accumulation in cecum caused by XOs fermentation leads to acidic pH of cecum.

Table 2: Final body weight, cecal total and wall weight and cecal pH of rat groups fed on experimental diets*

	Xo	G1	G2	G3	G4
Final body weight (g)	245±16[a]	260±19[b]	263±15[b]	273±20[c]	275±18[c]
Cecum weight (g)	3.46±0.45[a]	7.75±1.12[c]	4.69±0.65[b]	6.15±0.93[c]	6.29±0.88[c]
Cecal wall weight (g)	0.71±0.11[a]	1.45±0.08[c]	1.04±0.04[b]	1.10±0.01[c]	1.09±0.03[c]
Cecal content weight (g)	2.75±0.31[a]	6.30±1.57[c]	3.65±1.17[b]	5.05±1.60[c]	5.20±1.51[c]
Cecal pH	7.11±0.09[b]	5.95±0.10[a]	6.29±0.11[a]	6.18±0.10[a]	6.11±0.09[a]

*Values are given as means ± SEM

[abc]Means in the same row not sharing superscript letters differ significantly ($p<0.05$)

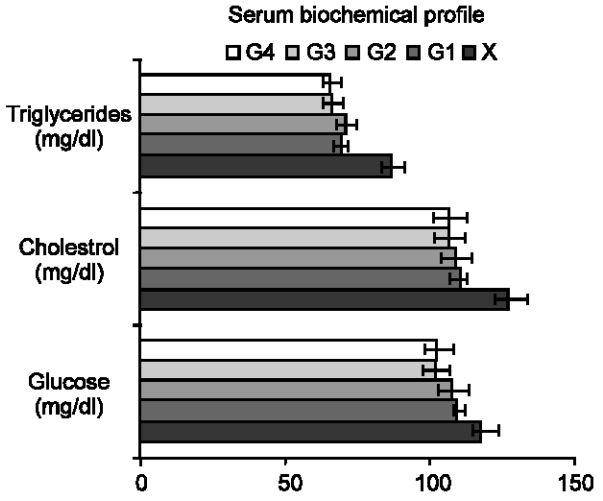

Fig. 1: Serum biochemical profile of rat groups fed to different experimental diets. Values shown are mean with standard error bars

Serum biochemical profile of the experimental groups is exhibited in Fig. 1. It was observed that XOs significantly lowered the serum glucose levels of rats fed on XOs enriched yogurt. Hence this trend was not observed in rats fed on plain yogurt. These results match with that of Imaizumi *et al.* (1991). As XOs are low pH resistant and can escape from digestion in the stomach and small intestine, so their malabsorption through digestion process helps to lower serum glucose levels. Some of Oligosaccharides (OS) have been reported to cause lowering serum glucose levels; fructo-oligosaccharides have been revealed to lessen postprandial glycemia and insulinemia in rats (Roberfroid and Delzenne, 1998). The exact mechanism(s) of this phenomenon is not clear yet; though, it is considered to be intervened by slowing gastric emptying, or altered hepatic metabolism. Tomomatsu (1994) in a study proposed the effective daily doses of few oligosaccharides (pure form) in humans including FOS (3.0 g) and XOs (0.7 g), suggestive of that XOs might be more effective than FOS in improving health regarding serum biochemical profile. Mean values of serum cholesterol level in all experimental groups is shown in Fig. 1. Yet, the reduction was not significant but a trend of lowering levels can be observed. Every day intake of oligosaccharides has also been reported to lower total serum cholesterol in humans by 0.226-0.566 mmol/L (Hidaka *et al.*, 1986). Fiordaliso *et al.* (1995) established modified hepatic lipid synthesis during inulin intake in a study on rats. Akalin *et al.* (1997) found that rats fed on acidophilus yogurt for 28 days have significantly lower ($p<0.01$) cholesterol level than for the control group. This parameter depends on feeding the experimental diet for a longer period, but the study period was 21 days in present study so no significant change could be noted. Serum triglyceride levels were lowered when compared to control group but no significant difference among other four groups was observed which is in accordance to the results of cholesterol level.

Conclusion: The present work concludes that XOs as prebiotic NDO and XOs enriched yogurt as synbiotic food play a definite role in altering serum biochemical profile of rats. Dietary supplementation with XOs leads towards microbial fermentation and lowers the cecal pH, therefore exerts beneficial health effects. The enrichment of yogurt with XOs established even better results. Further studies to draw firm conclusions should include efficacy study with humans.

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