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The effects of probiotics in the blood chemistry of Persian shepherd dogs

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Abstract

Probiotic therapy has been recommended for the treatment or prevention of a variety of conditions in different species. The benefits of probiotic consumption in human beings have been recognized for centuries. However, their application and efficacy in domestic animals hasonly recently been investigated. The aim of this study was to evaluate the efficacy of probiotic administration on the blood chemistry of Persian shepherd dogs. Ten male Persian shepherd dogs were divided into two groups. The control group was fed a control diet; whereas the treatment group was fed a control diet supplemented with probiotics (9 gr/10kg of diet). Several blood proteins were evaluated at multilple time-points during the study. The results showed insignificant changes in α_1 globulin ($P>0.05$), however α_2 globulin showed a significant decrease ($P<0.05$). Overall, in the total hemograms of both groups significant changes were observed in WBC, lymphocyte and neutrophil ($P<0.05$). The results indicate that oral administration of probiotic can improve the function of immune system.

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Introduction

Probiotics, as defined by the Food and Agriculture Organization of the United Nations (FAO) and World Health Organization (WHO), contain live microorganisms which, when administered in adequate amounts, have a health benefit for the host. probiotics are microbial supplements which can improve host body by microbial balance of intestine (FAO, 2006; Khani and Hosseini, 2008). A variety of microorganisms, typically lactic acid bacteria (LAB), such as *lactobacilli*, *bifidobacteria*, and *enterococci*, have been evaluated as potential probiotics. A small number of yeasts have also been evaluated. These bacteria generally produce lactic acid as their major end product. Lactic acid bacteria lack a terminal electron transport chain, therefore they are strictly fermentative, and catalase negative (McFarland *et al*., 1994; Filho Lima *et al*., 2000).

Probiotic therapy is becoming increasingly popular in veterinary medicine; however, few studies are available, particularly for dogs and cats. Probiotic therapy has been recommended for the treatment or prevention of a variety of conditions in different species (Weese, 2002). A number of probiotic products are commercially available for dogs and cats; they are available in tablet, capsule, paste, liquid or food supplements forms for daily administration (Hamilton Miller and Shah, 2002).

The beneficial properties include the ability to reduce antibiotic use and the positive perception of natural or alternative therapies. Not only antibiotic therapy is effective in reducing or eliminating pathogenic microorganisms, but also it is effective in eliminating the host's gut microflora, upsetting the natural microbial balance essential to a functional gastrointestinal tract and immune system. Among all the strains identified, best results were obtained with *Lactobacillus rhamnosus*, *Enterococcus faecium* and *Saccharomyces cerevisiae* as far as regards growth rates, pH and bile salts tolerance. Moreover, the identification profiles of microorganisms showed a better reliability for the products containing a single species whereas the ones composed of different strains were usually not satisfactory (Canganella *et al*., 1997).

Recently, several studies were conducted to demonstrate the benefits of probiotics in animals and human. Weese and Anderson (2002) indicated the ability of *Lactobacillus rhamnosus* strain (LGG) to save gastrointestinal transit and efficacy of this probiotic in the treatment of canine disease (Weese and Anderson, 2002).

According to the previous literature, this study was designed to determine the ability and effects of probiotic in changing the blood chemistry of Persian shepherd dogs.

Materials and methods

Animals and experiment

The experiment was conducted on 10 male Persian shepherd dogs ranging in age between one to two years. They were divided in two groups (n=5). The average body weight of each group was very similar (20-25 kg). The control and treatment groups were fed by healthy food and healthy food containing probiotics treatment containing Lactobacillus, Bifidobacterium and Enterococcus (9 gr/10kg of food; Perimalac, USA), respectively. Based on the recorded bodyweight of each animal, the following week's dry food ration was calculated and pre-weighed for each individual. All necessary vaccinations were administered during the 8-week period and the overall well-being of the animals was monitored by an in-house veterinarian.

Sample collection and processing

Blood samples were collected from lateral saphenous vein of each dog at the beginning of the trial and the same process was repeated at day 4, 9, 14 and 19. Five mL of blood sample with EDTA from each dog was dispensed into individual blood collection vials to be analyzed by Antech Diagnostics (Irvine, CA) for a complete blood count (CBC) and biochemical panel.

Aliquots of one mL were kept into appropriate tubes (tubes containing EDTA) designated by Antech Diagnostics to be used for a complete blood count and biochemical panel. Blood samples were analyzed using an Olympus Model AU 5400 System (Center Valley, PA). The biochemical panel was examined for the total protein, albumin and globulins.

Statistical analysis

The CBC and biochemistry panel data were analyzed using a repeated measures analysis of variance (ANOVA) using SAS v9.1 (SAS 2003). The first-order autoregressive covariance structure was deemed appropriate. The Slice effect was used to test any differences between treatments at various time points. Differences were considered significant at P<0.05 level. Pair-wise differences did not need to separate Least Squares Means because there were only two treatments.

Results

In recent years the use of probiotics has become a promising alternative to the utilization of antibiotics for the treatment and prevention of infections and for growth promotion as components of food for production and companion animals. This study showed clear effects of oral administration of probiotics on both immune response and improvement of serum biochemistry.

Table 1. The mean and standard deviation (SD) of total protein changes in the control and treatment groups.

Groups	Total protein of blood during the study (gr/lit)				
	Sampling days				
	24 hours ago	4	9	14	19
Control groups	81.45±9.97	78.6±9.75	74.7±0.98	78.18±10.34	78.2±0.56
Treatment groups	80.05±3.74	65.7±6.78	67.3±5.32	70.25±4.17	73.5±1.13

Statistical analysis of the interaction between each groups and Sampling days and total protein level, these changes do not differ significantly (P>0.05).

Table 2. The mean and standard deviation (SD) of Albumin changes in the control and treatment groups.

Groups	Albumin changes of blood during the study (gr/lit)				
	Sampling days				
	24 hours ago	4	9	14	19
Control groups	34.4±9.05	35.05±2.90	32.3±1.13	31.55±3.18	34.9±2.26
Treatment groups	36.25±1.9	33.1±4.66	32.7±3.95	27.25±1.48	30.06±2.74

Statistical analysis of the interaction between each groups and Sampling days and total Albumin level, these changes do not differ significantly (P>0.05).

In this study, the average total protein in the treatment group decreased from 80.05 to 65.7 during the first week which was not significant (P>0.05), but increased afterwards (Table 1). The average serum albumin showed a decrease in the treatment group which was not significant either (P>0.05) (Table 2). In serum globulins, α_1 globulin changes were not significant (P>0.05) (Table 3), but In the treatment group, α_2 globulin showed a significant decrease (P<0.05) (Table 4).

Table 3. The mean and standard deviation (SD) of α_1 globulin changes in the control and treatment groups.

Groups	α_1 globulin changes of blood during the study (gr/lit)				
	Sampling days				
	24 hours ago	4	9	14	19
Control groups	2.65±0.07	2.25±0.35	1.9±0.0	3.3±1.83	3.7±0.56
Treatment groups	3.45±0.77	2.35±0.77	2.95±0.21	2.6±0.84	2.95±0.63

Statistical analysis of the interaction between each groups and Sampling days and α_1 globulin level, these changes do not differ significantly (P>0.05).

In the treatment group the changes of β_1 globulin were significant (Table 5) and In the treatment group in β_2 globulin there was an increase which was significant ($P<0.05$) (Table 6). In the treatment group, serum Gamma globulin showed an increase during the research which was significant ($P<0.05$) (Table 7). Thus, in the total hemograms of both groups, significant changes were observed in WBC and their subgroups including lymphocyte and neutrophil ($P<0.05$). Hematocrit, hemoglobin and RBC changes in both groups were not significant ($P>0.05$).

Table 4. The mean and standard deviation (SD) of α_2 globulin changes in the control and treatment groups.

Groups	α_2 globulin changes of blood during the study (gr/lit)				
	Sampling days				
	24 hours ago	4	9	14	19
Control groups	11.65±3.88	11.65±0.21	11.05±2.47	10.6±4.1	10.35±0.77
Treatment groups	$10.95±0.77^{a}$	$8.1±0.56^{ab}$	$7.3±1.41^{ab}$	$6.1±1.27^{b}$	$6.7±1.69^{b}$

a, ab, b: values and means in column of treatment groups, these different superscripts differ significantly ($P<0.05$).

Table 5. The mean and standard deviation (SD) of β_1 globulin changes in the control and treatment groups.

Groups	β_1 globulin changes of blood during the study (gr/lit)				
	Sampling days				
	24 hours ago	4	9	14	19
Control groups	11.5±2.83	7.1±0.84	8.4±1.55	7.75±0.91	11.85±0.21
Treatment groups	$11.45±0.91^{a}$	$2.95±1.48^{d}$	$6.15±0.35^{c}$	$7.95±0.21^{bc}$	$9.5±0.7^{b}$

a, d, c, bc, b: values and means in column of treatment groups, these different superscripts differ significantly ($P<0.05$).

Discussion

Recently, several studies were conducted to demonstrate the benefits of probiotics in animals and human, most of which are in agreement with the results of the present study. Weese and Anderson (2002) indicated the ability of *Lactobacillus rhamnosus* strain (LGG) to save gastrointestinal transit and efficacy of this probiotic in the treatment of canine disease (Weese and Anderson, 2002). LGG has also been used successfully in the treatment of rotavirus-induced diarrhea in human (Pant *et al.*, 2007) as well as in the prevention or treatment of atopic diseases (Isolauri *et al.*, 1993; Viljanen *et al.*, 2005). It is interesting that even though LGG has been effective in different immune-mediated diseases, it induced only moderate cytokine production compared to the other strains in the present study.

Table 6. The mean and standard deviation (SD) of β_2 globulin changes in the control and treatment groups.

Groups	β_2 globulin changes of blood during the study (gr/lit)				
	Sampling days				
	24 hours ago	4	9	14	19
Control groups	10.3±1.34	12.35±2.89	11.05±0.91	11.25±2.19	9.05±0.35
Treatment groups	$7.95±0.49^{b}$	$7.85±0.35^{b}$	$8.35±1.06^{b}$	$12.1±1.69^{a}$	$12.5±0.84^{a}$

b, a: values and means in column of treatment groups, these different superscripts differ significantly ($P<0.05$).

The effect of probiotics on the immune response has been comprehensively reviewed (MacFarlane and Cummings, 2002; McNaught *et al.*, 2005). The majority of evidence from in vitro systems, animal models and humans suggest that probiotics can enhance both specific and nonspecific immune responses. These effects are believed to be mediated through activating macrophages, increasing cytokine levels, increasing natural killer cell activity and/or increasing levels of immunoglobulins (Ouwehand *et al.*, 2002).

Table 7. The mean and standard deviation (SD) of Gamma globulin changes in the control and treatment groups.

Groups	Gamma globulin changes of blood during the study (gr/lit)				
	Sampling days				
	24 hours ago	4	9	14	19
Control groups	8.3±0.91	9.05±3.88	10±0.98	10.9±2.96	8.35±1.34
Treatment groups	7.05±0.07[b]	10.9±1.55[ab]	9.6±0.98[ab]	12.2±1.69[a]	11.8±0.42[a]

b, ab, a: values and means in column of treatment groups, these different superscripts differ significantly (P<0.05).

Perelmuter *et al.,* (2008) isolated and identified *Lactobacillus spp.* from feces of a healthy dog in order to evaluate possible use of probiotics for dogs. The isolates survived in different pH and bile salts conditions, inhibited the *in vitro* growth of *Escherichia coli* and *Clostridium perfringens*, and adhered to intestinal mucus (Perelmuter *et al.*, 2008). In summary, the present study showed that, probiotics have an effective role in the reduction of systematic inflammation and increasing serum immunoglobulins, and that they cause positive changes in the function of humoral system and blood cells. Therefore, their addition to the diet of the animals can enhance the function of immune system.

References

Canganella F, Paganini S, Ovidi M, Vettraino AM, Bevilacqua L, Massa S, Trovatelli LD. 1997. A microbiological investigation on probiotic pharmaceutical products used for human health. Microbiological Research **152,** 171-179.

FAO Food, Nutrition Paper. 2006. Probiotics in food. Health and nutritional properties. FAO Food and Nutrition Paper 85. Rome, Italy.

Filho Lima JVM, Vieira EC, Nicoli JR. 2000. Antagonistic effect of Lactobacillus acidophilus, Saccharomyces boulardiiand Escherichia colicombinations against experimental infections with Shigella flexneriand Salmonella enteritidissubsp. typhimuriumin gnotobiotic mice. Journal of Applied Microbiology **88,** 365-370. http://dx.doi.org/10.1046/j.1365-2672.2000.00973.x

Hamilton Miller JMT, Shah S. 2002. Deficiencies in microbiological quality and labelling of probiotic supplements. International Journal of Food Microbiology **72,** 175-176.

Isolauri E, Majamaa H, Arvola T, Rantala I, Virtanen E, Arvilommi H. 1993. Lactobacillus caseistrain GG reverses increased intestinal permeability induced by cow milk in suckling rats. Gastroenterology **105,** 1643-50.

Khani MM, Hosseini SM. 2008. Effect of supplemented Lactobacillus acidophilus and Lactobacillus casei diet or water on serum cholesterol and triglyceride and performance of broilers1st Mediterranean Summit of WPSA. 22nd of February 2008. Thessaloniki. Book of Proceeding, 740-744.

MacFarlane GT, Cummings JH. 2002. Probiotics, infection and immunity. Current Opinion in Infectious Diseases **15,** 501-506.

McFarland LV, Surawicz CM, Greenburg RN, Fekety R, Elmer GW, Moyer KA, Melcher SA, Bowen KE, Cox JL, Noorani Z, Harrington G, Rubin M, Greenwald D. 1994. A randomized placebo-controlled trial of Saccharomyces boulardiiin combination with standard antibiotics for Clostridium difficiledisease. Journal of the American Medical Association **271,** 1913-1918. http://dx.doi.org/10.1001/jama.1994.03510480037031.

McNaught CE, Woodcock NP, Anderson AD, Mac-Fie J. 2005. A prospective randomised trial of probiotics in critically ill patients. Clinical Nutrition **24,** 211-219. http://dx.doi.org/10.1016/j.clnu.2004.08.008.

Ouwehand AC, Salminen S, Isolauri E. 2002. Probiotics: an overview of beneficial effects. Antonie van Leeuwenhoek **82,** 279-289. http://dx.doi.org/10.1023/A:1020620607611.

Pant N, Marcotte H, Brussow H, Svensson L, Hammarstrom L. 2007. Effective prophylaxis against rotavirus diarrhea using a combination of Lactobacillus rhamnosus GG and antibodies. BMC Microbiology **7,** 86. http://dx.doi.org/10.1186/1471-2180-7-86.

Perelmuter K, Fraga M, Zunino P. 2008. In vitro activity of potential probioticLactobacillus murinus isolated from the dog. Journal of Applied Microbiology **104,** 1718-1725. http://dx.doi.org/10.1111/j.1365-2672.2007.03702.x. Epub 2008 Jan 9.

Viljanen M, Savilahti E, Haahtela T, Juntunen-Backman K, Korpela R, Poussa T, Tuure T, Kuitunen M. 2005. Probiotics in the treatment of atopic eczema/dermatitis syndrome in infants: a double-blind placebo-controlled trial. Allergy **60,** 494-500. http://dx.doi.org/10.1111/j.1398-9995.2004.00514.x.

Weese JS, Anderson MEC. 2002. Preliminary evaluation of Lactobacillus rhamnosus strain GG, a potential probiotic in dogs. Canadian Veterinary Journal **43,** 771-774.

Weese JS. 2002. Microbiologic evaluation of commerical probiotics. Journal of the American Veterinary Medical Association **220,** 794-797.