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Assessment of blood metabolites, serum enzymes, and serum minerals in dietary probiotics fed captive Asian elephants

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Abstract: This study was organised to evaluate the effect of supplementation of dietary probiotics on blood biochemistry in 18 captive Asian elephants of 30-62 years age and 3495 ± 133.34 kg body weight (BW) for two months. To observe the results in different groups, i.e. control with no probiotic (T_1) and the other two with probiotics which contained 1×10^9 CFU/g concentrate of *Lactobacillus acidophilus* (T_2) and 1×10^9 CFU/g concentrate of *Saccharomyces cerevisiae* (T_3), they were given 50 kg BW/day (dose range 48-89 g per day/elephant). The elephants were randomly divided into three groups with six elephants each. They were fed on *ad libitum* green fodder. Blood biochemical constituents namely: (i) blood metabolites like glucose, total protein, albumin, cholesterol, creatinine, and blood urea nitrogen, (ii) serum enzymes like aspartate transaminase, alanine transaminase, and alkaline phosphatase, and (iii) serum minerals like calcium and phosphorus were analysed at the end of research to evaluate the physiological impacts of nutrients in the elephant's body. It didn't establish any health problem to the elephants since the parameters determined in the blood were observed well within the normal physiological range, and no significant differences were noticed due to the effect of treatment. It is concluded that blood biochemistry remained unaffected on the supplementation of probiotics in Asian elephants.

Key words: Asian elephant, blood biochemistry, physiological impact, probiotics

1. Introduction

The Asian elephant (*Elephas maximus*) is a monogastric herbivore, and its digestive system is comparable to that of equids [1], rabbits, and rodents. The elephant's large fermentative chambers harbour diverse anaerobic microbes consisting of bacteria, fungi, and protozoa for the fermentation and breakdown of fibrous feed materials [2]. The intestinal microorganisms have a vast impact on the gut health and performance of elephants and their vital aspects in wildlife health care, welfare management, and conservation programs.

Amongst of nutraceuticals, probiotics are the live microbial cultures that have been extensively used in animals because of their ability to modulate intestinal microorganism and immune system. The inclusions of probiotics furnish health advantages: improve growth performance and nutrient digestibility to the host when supplemented in adequate dosage [3]. It also lessens serum cholesterol and the incidence of diarrhea in livestock [4,5].

Appraisement of the health and nutritional status of livestock is inestimable in present-day animal husbandry. Blood biochemical investigation is the right way of assessing the health status of livestock used in various feed trials as it plays a significant role in the nutritional, physiological, and pathological state of animals. Serum concentrations of metabolites such as total proteins, glucose, globulin, albumin, cholesterol, blood urea nitrogen, creatinine, and minerals indicate the degree of the metabolism of proteins, energy, and other nutrients in the livestock [6,7]. Variations in the concentration of circulating nutrient-sensitive metabolites are essential signals of the livestock's metabolic status and the activity of the organs [8,9]. Factors including the physiological condition of livestock, nutrition, health status, breed, age, and season may vary the concentration of these metabolites in the serum [10]. In this context, Olorode & Longe [11] have summarized that nutrition plays an important role and intervenes with the serum metabolites and other constituents' concentration levels. Therefore, a study was determined to assess the impact of administration of probiotics on the serum biochemical profile of captive Asian elephants.

2. Materials and methods

2.1. Study animals and design

The study was organised from August to October 2019 at Elephant Village, Jaipur (India) (26° 59'47" N and 75°

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52'35" E). Eighteen adult captive female Asian elephants of 30-62 years age, having similar body weight (3495 ± 133.34 kg), and of uniform conformation were randomly selected. They were divided into three similar groups with six elephants each. The elephants were stall-fed a consistent diet of green pearl millet fodder as basal diet throughout the research period, i.e. for a period of two months including ten days for adaptability with basal diet and fifty days for probiotics feeding. They were also allowed for a routine elephant safari. Fresh and clean *ad libitum* water was provided to all the elephants. A prophylaxis dose of broad-spectrum anthelmintic, i.e. Panacur Bolus (fenbendazole) as 5mg/kg body weight, was given to all the elephants prior to experimental feeding of probiotics.

2.2. Probiotics

The probiotics, i.e. *Lactobacillus acidophilus* and *Saccharomyces cerevisiae* were supplemented along with basal diet during the feeding trial of 50 days to all the elephants in T_2 and T_3 treatment groups, respectively. The probiotic supplemented in this study was a commercially available powder product by Meteoric Biopharmaceuticals Pvt. Ltd. (Gujarat, India). The products contained 1×10^9 colony-forming units (CFU)/g concentrate of *Lactobacillus acidophilus* and 1×10^9 colony-forming units (CFU)/g concentrate of *Saccharomyces cerevisiae.* One gram probiotic was given per 50 kg body weight orally with jaggery, whereas the only jaggery was supplemented as a placebo along with the basal diet to the elephants of T_1 i.e. control group. The horse was considered as a model animal for calculating probiotic requirements and designing a diet for elephants [1].

2.3. Blood collection and analysis

For the analysis of blood metabolites, serum enzymes, and serum minerals, five mL of blood was collected after the end of the feeding trial between 07:00-09:00 AM from all elephants by puncturing ear vein. The blood was taken directly into a vacutainer tube and transported to the laboratory on ice, where serum was harvested and tested on the same day. From the serum samples, (i) blood metabolites like glucose, total protein, albumin, cholesterol, creatinine, and blood urea nitrogen (ii) serum enzymes like aspartate transaminase (AST), alanine transaminase (ALT), and alkaline phosphatase (ALP), and (iii) serum minerals like calcium and phosphorus were analysed using the commercial kits on fully automated biochemistry analyser "Turbochem100" (Awareness Inc., Connecticut, USA) as per manufacturer's protocol. Each sample was labelled with a unique identity (ID) number, date, and time of sampling.

2.4. Statistical analysis

All statistical analysis of data was performed using SPSS 16 version (IBM Corp., Armonk, NY, USA) for windows. The difference among groups was determined by one-way ANOVA analysis. The significant effects of different means were compared by Duncan's new multiple range test [12]. Significance was defined at $P < 0.05$. All values represent mean ± standard errors of the mean.

2.5. Ethical considerations

All applicable national, international, and institutional guidelines for the care and handling of animals were followed during blood sample collection. The study was conducted with the prior approval of the Additional Principal Chief Conservator of Forest and Chief Wildlife Warden, Rajasthan, Jaipur, India (Ref. no. 1159, dated May 28, 2019) and the Government of India, Ministry of Environment, Forest and Climate Change, Wildlife Division, New Delhi (Ref. no.1-22/2019 WL, dated October 16, 2019).

3. Results and discussion

The animals were in excellent condition throughout the study period without signs of abnormal health. Supplementation of probiotics, i.e. *Lactobacillus acidophilus* and *Saccharomyces cerevisiae* for 50 days did not affect their health. Serum biochemical analysis is used to determine liver, heart, and kidney functioning and is mostly used as a signal of the physiological effects of nutrients in the body of animals.

The data that were enumerated during the investigation of serum biochemistry parameters namely: (i) blood metabolites like glucose, total protein, albumin, cholesterol, creatinine, and blood urea nitrogen, (ii) serum enzymes like aspartate transaminase (AST), alanine transaminase (ALT), and alkaline phosphatase (ALP), and (iii) serum minerals like calcium and phosphorus are shown in Table.

3.1. Blood metabolite

The mean values of serum glucose observed in this investigation were found well within the normal physiological range of 25.2-102.6 (mg/dL) as reported in Asian elephants [13,14,15]. The mean value of glucose was observed lowest in T_3 and highest in T_1 groups. However, the supplementation of probiotics did not have any effect on glucose among the groups. A similar nonsignificant effect, supporting the result of the present investigation, has also been noticed earlier in rabbits [16,17,18,19,20]. In another study, elevated ($P < 0.05$) serum glucose level was also reported in *Saccharomyces cerevisiae* fed pigs [21].

The mean values of total protein obtained in the present investigation for different groups were within the physiological range of 7.2-10.2 (g/dL) [13] and it attests to the results of earlier studies conducted in Asian elephants [22,14,23]. The statistical analysis of data of total protein revealed a nonsignificant effect among the treatments. The results are consistent with earlier findings [24,20], which indicate that serum level of total protein remained the same on the administration of probiotics in lamb and

Table. Different blood biochemical constituents in probiotics fed Asian elephants.

Parameters (Mean ± SEM)	T_1	T_2	T_3	P-value
Blood metabolites				
Glucose (mg/dL)	31.16 ± 6.11	29.58 ± 9.18	26.65 ± 8.25	0.92
Total Protein (g/dL)	8.12 ± 0.38	8.12 ± 0.41	8.24 ± 0.47	0.97
Albumin (g/dL)	3.05 ± 0.15	3.18 ± 0.15	3.08 ± 0.19	0.83
Cholesterol (mg/dL)	33.25 ± 3.22	31.08 ± 3.01	30.60 ± 5.73	0.89
Creatinine (mg/dL)	1.30 ± 0.07	1.24 ± 0.10	1.25 ± 0.06	0.87
Blood Urea Nitrogen (mg/dL)	8.26 ± 0.17	8.18 ± 0.09	8.26 ± 0.09	0.87
Serum enzymes				
AST (IU/L)	24.68 ± 1.25	23.75 ± 1.74	27.22 ± 1.77	0.32
ALT (IU/L)	2.07 ± 0.48	3.37 ± 0.48	2.28 ± 0.32	0.11
Alkaline Phosphatase (ALP)(IU/L)	54.13 ± 6.77	59.67 ± 7.90	58.74 ± 3.60	0.81
Serum minerals				
Calcium (mg/dL)	11.53 ± 0.29	11.10 ± 0.62	11.42 ± 0.36	0.78
Phosphorus (mg/dL)	4.95 ± 0.48	6.47 ± 0.62	6.26 ± 0.46	0.12

T_1- Control, T_2- probiotic (*L. acidophilus)*, and T_3- probiotic (*S. cerevisiae)*.

rabbit, respectively. However, Onifade et al. [25] revealed that serum protein level was elevated with dietary yeast in rabbits and highly significant (P < 0.001) with *Lactobacillus* spp. in weaned piglets [26,27].

The statistical analysis of data of serum albumin revealed a nonsignificant effect among the three groups. The results are in accordance with the earlier observations [24,20] in lamb and rabbit, respectively. However, Onifade et al. [25] revealed that albumin level was increased with dietary yeast in rabbit, *Lactobacillus* spp. in piglets [26,27] and *Bacillus* spp. in lambs [28]. The albumin level obtained in the present investigation for different groups was found to be well within the physiological range of 2.6-3.6 (g/dL) reported in Asian elephants [13,14]. However, it was found towards a slightly higher side as compared to the results observed in earlier studies [22,29].

The statistical analysis of data of serum cholesterol revealed a nonsignificant effect among the three groups. The mean values of the present study are in corroboration with the earlier findings as observed in broiler chicken [30] and lamb [24], whereas it exhibits significant differences with the results obtained in other studies in rat [31] and in mice [32]. Similarly, earlier studies [25,33] reported that serum level of cholesterol was decreased with dietary inclusion of yeast and *Lactobacillus* spp. in rabbits, respectively. The mean cholesterol level obtained in the present investigation for different groups was found to be well within the normal reference range of 8.1-51.66 (mg/dL) [13]; however, it was found towards the lower side as compared to the data observed in earlier studies [22,29,14]. It was found lowest in T_3 and highest in T_1 (control) groups.

The mean values of creatinine obtained in the present investigation for different groups were found to be well within the normal physiological range of 0.9-1.8 (mg/dL) [23] and it attests to the earlier observations in Asian elephants [22,29,14,15]. The statistical analysis of data of creatinine revealed a nonsignificant effect among the three groups. The study was in agreement with earlier findings in weaned piglets [26]. However, its concentration was decreased significantly in probiotic fed large felids [34] and rats [35].

The blood urea nitrogen concentration obtained in the present investigation for different groups was found to be well within the normal range of 4.2-19.7 (mg/dL) [23] and the mean values are in corroboration with the earlier findings in Asian elephants [22,29,14,15]. Supplementation of probiotics did not have any effect (P > 0.05) on blood urea nitrogen among the groups. Similar results were reported in ewes [36], large felids [34] and weaned piglets [26]. However, a significantly decreased concentration of blood urea nitrogen was observed in probiotic fed rats [35] and lambs [28].

The liver enzymes, i.e., serum AST, ALT, and ALP activities, were used to estimate liver functions. The increased concentration of these enzymes was associated with the degeneration of hepatic cells [37,38] and muscle tissues [39,40].

The statistical analysis of data of AST revealed a nonsignificant effect among the groups. The values are in corroboration with the findings reported earlier [41]. The serum level of AST was found to be reduced with the administration of dietary yeast in rabbits [25], multistrain probiotics in rats [31], and *Bacillus* spp. in lambs [28], whereas all the treatments (Kefir rich probiotic levels 3%, 6%, 9%, and 12%) significantly increased the AST enzyme level as compared to the control group ($P < 0.05$) in Japanese quails [42]. The AST level obtained in the present investigation for different groups was found to be well within the normal reference range of 10.1-39.6 U/L [23] and the mean values are consistent with earlier findings in Asian elephants [22,14]. The mean AST value of the present study was found to be highest in the T_3 group.

The statistical analysis of data of ALT enzyme in both treatment groups was more than the control group, but the variations were insignificant, and the mean values are in accordance with the earlier studies in broiler chicken [41] and Japanese quails [42]. On the other hand, serum level of ALT was decreased with dietary yeast in rabbits [25], multistrain probiotics in rats [31], and *Bacillus* spp. in lambs [28]. However, the ALT level observed in the present investigation for different groups was found to be well within the average range of 0-5.6 U/L, as reported in Asian elephants [23].

The ALP level observed in the present study for the different groups was found to be well within the average reference range of 0-281.5 U/L [23] and it attests to the results of earlier studies conducted in elephants [22,14]. The serum concentrations of ALP of the present study were not affected by dietary treatments and are in agreement with earlier reports in ewes [36] and weaned piglets [26]. However, serum levels of ALP were decreased in rabbits [25] and Japanese quails [42] treated with the low levels of probiotics.

3.2. Serum minerals

Limited information is available on the serum minerals in response to dietary probiotic administration. Macro and micro minerals are advantageous to livestock and act as structural and functional cofactors in metal-containing enzymes. These minerals play a vital role in livestock's physiology [43].

The mean serum calcium level obtained in the present investigation for the different groups was found to be slightly lower than the normal reference range of 31.14-56.34 mg/dL [44] and 18.10 ± 1.78 mg/dL [45]. The statistical analysis of data of serum calcium revealed a nonsignificant effect among the groups. The present results are in agreement with the findings reported in pigs [46], broiler chicks [47] and rats [48]. However, increased and significantly decreased ($P \leq 0.001$) serum calcium levels were reported in rabbits [49,50] and lambs [28], respectively.

The statistical analysis of data of serum phosphorus revealed an insignificant increase in T_2 and T_3 groups as compared to the control group. These results are in agreement with earlier findings in cows [51]. However, a significant increase in serum phosphorus level was seen in rabbits [50] and lambs [28]. The serum phosphorus level recorded in the present investigation for different groups was observed to be in corroboration with the earlier studies [45] but found to be a little lower than the data cited in Asian elephants [44,13]. The mean phosphorus value was found to be lowest in the diet of the T_1 group. However, the Ca/P ratio was found to be well within the normal reference range [13].

4. Conclusion

It is concluded that the administration of dietary probiotics *Lactobacillus acidophilus* and *Saccharomyces cerevisiae* to Asian elephant did no significant changes in blood metabolites, serum enzymes, and serum minerals. It didn't establish any health problem to the elephants since the parameters determined in the blood under the study conditions were observed well within the normal physiological range of a healthy animal.

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References

1. Clauss M, Loehlein W, Kienzle E, Wiesner H. Studies on feed digestibilities in captive Asian elephants (Elephas maximus). Journal of Animal Physiology and Animal Nutrition 2003; 87: 160-173.

2. Clements ET, Maloiy GMO. The digestive physiology of three East African herbivores: the elephant, rhinoceros and hippopotamus. Journal of Zoology (London) 1982; 198: 141-156.

3. Alayande KA, Aiyegoro OA, Ateba CN. Probiotics in animal husbandry: applicability and associated risk factors. Sustainability 2020; 12: 1087. doi:10.3390/su12031087

4. Zhao P, Kim I. Effect of direct-fed microbial on growth performance, nutrient digestibility, fecal noxious gas emission, fecal microbial flora and diarrhea score in weanling pigs. Animal Feed Science and Technology 2015; 200: 86-92.

5. Lan R, Tran H, Kim I. Effects of probiotic supplementation in different nutrient density diets on growth performance, nutrient digestibility, blood profiles, fecal microflora and noxious gas emission in weaning pig. Journal of the Science of Food and Agriculture 2017; 97: 1335-1341.

6. Pambu-Gollah R, Cronje PB, Casey NH. An evaluation of the use of blood metabolite concentrations as indicators of nutritional status in free-ranging indigenous goats. South African Journal of Animal Science 2000; 30: 115-120.

7. Agenas S, Heath MF, Nixon RM, Wilkinson JM, Phillips CC. Indicators of under-nutrition in animal. Animal Welfare 2006; 15: 149-160.

8. Lindsay DB, Hunter RA, Gazzola C, Spiers WG, Sillence MN. Energy and growth. Australian Journal of Agricultural Research 1993; 44: 875-889.

9. Wettemann RP, Lents CA, Ciccioli NH, White FJ, Rubio I. Nutritional and suckling mediated an ovulation in beef cows. Journal of Animal Science 2003; 81 (2): 48-59.

10. Ndlovu T, Chimonyo M, Okoh AI, Muchenje V, Dzama K et al. Assessing the nutritional status of beef cattle: current practices and future prospects. African Journal of Biotechnology 2007; 6 (24): 2727-2734.

11. Olorode BR, Longe OG. Growth, nutrient retention, haematology and serum chemistry of pullet chicks fed shea butter cake in the humid tropics. Archivos De Zootecnia 1999; 49 (187): 444.

12. Steel RGD, Torrie JH, Dickey DA. Principles and Procedures of Statistics. A Biometrical Approach. 3rd ed. New York, USA: McGraw Hill Book Company; 1997.

13. Niemuller C, Gentry PA, Liptrap RM. Longitudinal study of haematological and biochemical constituents in blood of the Asian elephant (Elephas maximus). Comparative Biochemistry and Physiology 1990; 96A (1): 131-134.

14. Das A, Saini M, Katole S, Kullu SS, Swarup D et al. Effect of feeding different levels of wheat roti on nutrient utilization and blood metabolite profile in semi-captive Asian elephants (Elephas maximus). Journal of Animal Physiology and Animal Nutrition 2015; 99: 367-378.

15. Sahoo M. Management of foot lesions in Asian elephants (Elephas maximus) of Odisha. M.V.Sc. Orissa University of Agriculture and Technology, Odisha, 2015.

16. Simonová M, Marcinakova M, Strompfova V, Cobanova K, Gancarcikova S et al. Effect of probiotics Lactobacillus rhamnosus gg and new isolate Enterococcus faecium ef2019 (ccm 7420) on growth, blood parameters, microbiota and coccidia oocysts excretion in rabbits. International Journal of Probiotics and Prebiotics 2008; 3 (1): 7-14.

17. Seyidoğlu N, Galip N. Effects of Saccharomyces cerevisiae and Spirulina platensis on growth performances and biochemical parameters in rabbits. Kafkas Üniversitesi Veteriner Fakültesi Dergisi 2014; 20: 331-336. doi:10.9775/kvfd.2013.9988

18. Sarat Chandra A, Mahender M, Harikrishna C. Effect of dietary supplementation of probiotics and enzymes on the blood biochemistry and immune competence of rabbits reared under two housing systems. Livestock Research International 2015; 3 (1): 20-24.

19. Belhassen T, Bonai A, Gerencsér Zs, Matics Zs, Tuboly T et al. Effect of diet supplementation with live yeast Saccharomyces cerevisiae on growth performance, caecal ecosystem and health of growing rabbits. World Rabbit Science 2016; 24: 191-200. doi:10.4995/wrs.2016.3991

20. Kalma RP, Chauhan HD, Srivastava AK, Pawar MM. Growth and blood profile of broiler rabbits on probiotic supplementation. Indian Journal of Small Ruminants 2018; 24 (1): 66-69. doi: 10.5958/0973-9718.2018.00031.4

21. Kumar S, Verma AK, Mondal SK, Gupta M, Patil A et al. Effect of live *Saccharomyces cerevisiae* feeding on serum biochemistry in early weaned cross bred piglets. Veterinary World 2012; 5 (11): 663-666.

22. Katole S, Das A, Agarwal N, Prakash B, Saha SK et al. Influence of work on nutrient utilisation in semicaptive Asian elephants (Elephas maximus). Journal of Applied Animal Research 2014; 42 (4): 380-388. doi: 10.1080/09712119.2013.875904

23. Janyamethakul T, Sripiboon S, Somgird C, Pongsopawijit P, Panyapornwithaya V et al. Hematologic and biochemical reference intervals for captive Asian elephants (Elephas maximus) in Thailand. Kafkas Üniversitesi Veteriner Fakültesi Dergisi 2017; 23 (4): 665-669. doi: 10.9775/kvfd.2017.17380

24. Antunovic Z, Speranda M, Liker B, Seric V, Sencic D et al. Influence of feeding the probiotic Pioneer PDFM to growing lambs on performances and blood composition. Acta Veterinaria-Beograd 2005; 55: 287-300.

25. Onifade AA, Obiyan RI, Onipede E, Adejumo DO, Abu OA et al. Assessment of the effects of supplementing rabbit diet with a culture of "Saccharomyces cerevisiae" using growth performance, blood composition and clinical enzyme activities. Animal Feed Science and Technology 1999; 77: 25-32. doi:10.1016/S0377-8401(98)00244-2

26. Dong X, Zhang N, Zhou M, Tu Y, Deng K et al. Effects of dietary probiotics on growth performance, faecal microbiota and serum profiles in weaned piglets. Journal of Basic Microbiology 2013; 49 (2): 220-226.

27. Dowarah R, Verma AK, Agarwal N, Singh P, Singh BR. Selection and characterization of probiotic lactic acid bacteria and its impact on growth, nutrient digestibility, health and antioxidant status in weaned piglets. PLoS ONE 2018; 13: e0192978.

28. El-Sayed AA, Mousa SA. Effects of administration of probiotic on body growth and hematobiochemical profile in growing Barki lambs. Comparative Clinical Pathology 2019; 29: 297-303.

29. Allwin B, Senthil KG, Kalaignan Pa, Senthil NR, Vairamuthu S et al. Serum biochemistry of the Asian elephant. International Journal of Advanced Multidisciplinary Research 2015; 2 (4): 62-66.

30. Kwan OS, Kim IH, Hong W, Han YK, Lee SH et al. Effects of probiotics supplementation on growth performance, blood composition, and fecal noxious gas of broiler chickens. Korean Journal of Poultry Science 2002; 1: 1-6.

31. Mustari A, Ahmad N. Effects of probiotics on serum biochemical parameters in rats. The Bangladesh Veterinarian 2011; 28 (2): 70-74.

32. Salahuddin M, Akhter H, Akter S, Miah MA, Ahmad N. Effects of probiotics on haematology and biochemical parameters in mice. The Bangladesh Veterinarian 2013; 30 (1): 20-24.

33. El-deek AA, Albanoby MA, El-Naga MKA. Evaluation of super action probiotic as a natural growth promoter for growing rabbits. Egyptian Poultry Science Journal 2013; 33: 407-419.

34. McCain S, Allender MC, Schumacher J, Ramsay E. The effects of a probiotic on blood urea nitrogen and creatinine concentrations in large felids. Journal of Zoo and Wildlife Medicine 2011; 42 (3): 426-429. doi: http://dx.doi.org/10.1638/2010-0235.1

35. Wang F, Jiang YS, Liu F. The influence of mutant lactobacilli on serum creatinine and urea nitrogen concentrations and renal pathology in 5/6 nephrectomized rats. Renal Failure 2016; 38 (9): 1441-1447. doi: 10.1080/0886022X.2016.1227617

36. Mašek T, Mikulec Z, Valpotic H, Antunac N, Mikulec N et al. Influence of live yeast culture (*Saccharomyces cerevisiae*) on milk production and composition, and blood biochemical of grazing dairy ewes during the milking period. Acta Veterinaria Brno 2008; 77: 547-554.

37. Quezada T, Cuellar H, Valdiviaand AG, Reys JI. Effects of aflatoxin B1 on the liver and kidneys of broiler chickens during development. Comparative Biochemistry and Physiology Part C: Toxicology & Pharmacology 2002; 125: 265-272.

38. Fatemi F, Allameh A, Dadkhah A, Forouzadeh M, Kazemnejad S et al. Changes in hepatic cytosolic glutathione S-transferase activity and expression of its class-P during prenatal and postnatal periods in rats treated with aflatoxin B1. Archives of Toxicology 2006; 80: 572-579.

39. Ozyurt B, Iraz M, Koca K, Ozyurt H, Sahin S. Protective effects of caffeic acid phenethyl ester on skeletal muscle ischemiareperfusion injury in rats. Molecular Cell Biochemistry 2006; 292: 197-203.

40. Scholl PF, McCoy L, Kensler TW, Groopman GD. Quantitative analysis and chronic dosimetry of the aflatoxin B1 plasma albumin adduct Lys-AFB1 in rats by isotope dilution mass spectrometry. Chemical Research in Toxicology 2006; 19: 44-49.

41. Cenesiz S, Yaman H, Ozcan A, Kart A, Karademir G. Effects of Kefir as a probiotic on serum cholesterol, total lipid, aspartate amino transferase and alanine aminotransferase activities in broiler chicks. Medycyna Weterynaryjna 2008; 64: 168-170.

42. Vahdatpour T, Babazadeh D. The effects of kefir rich in probiotic administration on serum enzymes and performance in male Japanese quails. Journal of Animal and Plant Sciences 2016; 26: 34-39.

43. Shrivastava AK, Jha RR. Effects of different chemical composition and probiotic on the haematobiochemical profile of rabbit. Electronic Journal of Environmental, Agricultural and Food Chemistry 2010; 9: 1507-13.

44. Brown IRF, White PT. Elephant blood haematology and chemistry. Comparative Biochemistry and Physiology 1980; 65B: 1-12.

45. Gromadzka-Ostrowska J, Jakubów K, Zalewska B, Krzywicki Z. Haematological and blood biochemical studies in female domesticated Indian elephants (Elaphas maximus L.). Comparative Biochemistry and Physiology Part A: Physiology 1988; 89 (3): 313-315. doi:10.1016/0300-9629(88)91031-6

46. Kunavue N, Lien T. Effects of fulvic acid and probiotic on growth performance, nutrient digestibility, blood parameters and immunity of pigs. Journal of Animal Science Advances 2012; 2 (8): 711-721.

47. Hashemzadeh F, Rahimi S, Torshizi MA, Masoudi AA. Effects of probiotics and antibiotic supplementation on serum biochemistry and intestinal microflora in broiler chicks. International Journal of Agriculture and Crop Sciences 2013; 5 (20): 2394.

48. Soleymanzadeh MS, FathiZadeh S, Majidpour A, Mohammad N, Fagheei AZ et al. Have probiotic therapy effect on calcium and cholesterol serum levels in demyelinated hippocampus? Infection, Epidemiology and Microbiology 2017; 3 (3): 82-85.

49. Simonová MP, Lauková A, Chrastinova L, Stpová V, Faix S et al. Enterococcus faecium CCM7420, bacteriocin PPB CCM7420 and their effect in the digestive tract of rabbits. Czech Journal of Animal Science 2009; 54: 376-386.

50. Shah AA, Yuan X, Khan RU, Shao T. Effect of lactic acid bacteria-treated King grass silage on the performance traits and serum metabolites in New Zealand white rabbits (Oryctolagus cuniculus). Journal of Animal Physiology and Animal Nutrition 2017; 00: 1-7. https://doi.org/10.1111/jpn.12854

51. Jatkauskas J, Vrotniakienė V. Effect of L-plantarum, Pediococcus acidilactici, Enterococcus faecium and L-lactis microbial supplementation of grass silage on the fermentation characteristics in rumen of dairy cows. Veterinarija Ir Zootechnika 2007; 1: 40-62.