Kafkas Universitesi Veteriner Fakultesi Dergisi
ISSN: 1300-6045 e-ISSN: 1309-2251
Journal Home-Page: http://vetdergikafkas.org

Kafkas Univ Vet Fak Derg
27 (1): 111-115, 2021
DOI: 10.9775/kvfd.2020.24855

RESEARCH ARTICLE

Effects of Kefir on Blood Parameters and Intestinal Microflora in Rats: An Experimental Study

Bulent OZSOY [1,a(*)] Zafer CANTEKIN [2,b] Sakine YALCIN [3,c] Hamdullah Suphi BAYRAKTAR [4,d]

[1] Department of Animal Nutrition and Nutritional Disease, Faculty of Veterinary Medicine, Aydın Adnan Menderes University, TR-09016 Aydın - TURKEY
[2] Department of Microbiology, Faculty of Veterinary Medicine, Hatay Mustafa Kemal University, TR-31040 Hatay - TURKEY
[3] Department of Animal Nutrition and Nutritional Disease, Faculty of Veterinary Medicine, Ankara University, TR-06110 Ankara - TURKEY
[4] Department of Microbiology, Faculty of Medicine, Hatay Mustafa Kemal University, TR-31060 Hatay - TURKEY
ORCIDs: [a] 0000-0003-0045-3790; [b] 0000-0002-8837-0262; [c] 0000-0001-8640-2729; [d] 0000-0003-3400-9292

Article ID: KVFD-2020-24855 Received: 17.08.2020 Accepted: 14.12.2020 Published Online: 14.12.2020

Abstract

A probiotic product of kefir is widely consumed by human beings. The purpose of this research was to investigate the effects of kefir on blood parameters and intestinal flora in rats. A total of 24 female rats were used in this study. During 35 days of experimental period, rats were fed with a commercial diet and water was provided *ad libitum*. Kefir was given at the levels of 10 mL/kg, 20 mL/kg and 30 mL/kg with oral gavage to the first, second and third treatment groups, respectively. Kefir was not given to the control group. The number of yeast was found to be $1.65x10^7$ and the number of lactobacilli was found to be $4x10^8$ in kefir. At the end of the experiment, blood samples were taken from all rats. Blood plasma parameters and were investigated. The intestinal microflora was investigated by classical colony counting method. No differences were observed among the groups in total protein, albumin, uric acids, SGPT, SGOT, alkaline phosphatase and phosphorus in blood plasma. The plasma triglyceride and cholesterol levels in the second and third groups were lower than control group ($P<0.05$). No differences were observed in the intestinal pH levels among groups. Although total bacteria number of intestinal microflora was not different in groups, the number of Enterobactericeae and coliform bacteria in the third group was lower than the other groups ($P<0.001$). The number of Lactobacilli and the yeast level in the intestinal contents were increased by the usage of kefir ($P<0.001$). It was concluded that positive effects of the kefir were observed in intestinal microflora with increasing the number of beneficial bacteria and decreasing harmful bacteria and therefore kefir has a positive effect on the health of the animals.

Keywords: *Kefir, Rat, Performance, Blood parameters, Intestinal microflora*

Ratlarda Kefirin Bağırsak Mikroflorası ve Bazı Kan Parametrelerindeki Rolü: Deneysel Çalışma

Öz

Probiyotik bir ürün olan kefir insanlar tarafından yaygın olarak tüketilmektedir. Bu araştırmanın amacı, kefirin ratlarda kan parametreleri ve bağırsak florası üzerindeki etkilerini araştırmaktır. Bu çalışmada toplam 24 dişi rat kullanıldı. Otuzbeş günlük deney süresi boyunca, ratlar ticari rat yemi ile beslendi ve su *ad libitum* verldi. Kefir, deneme gruplarına sırası ile 10, 20 ve 30 mL/kg seviyelerinde oral gavajla verildi. Kontrol grubuna kefir verilmedi. Kefir maya sayısı $1.65x10^7$ lactobacilli sayısı $4x10^8$ bulunmuştur. Kan plazmasındaki total protein, albümin, ürik asit, SGPT, SGOT, alkalin fosfataz ve fosfor açısından gruplar arasında fark gözlenmedi. İkinci ve üçüncü gruptaki plazma trigliserid ve kolesterol, seviyeleri kontrol grubuna göre daha düşüktü ($P<0.05$). Gruplar arasında bağırsak pH seviyelerinde farklılık gözlenmedi. Bağırsak mikroflorasında total bakteri sayısı kontrol ve deneme gruplarında fark çıkmazken. Üçüncü deneme grubunda, enterobakteri ve koliform bakteri sayısı diğer gruplardan daha düşük ($P<0.001$) bulunmuştur. Aynı şekilde Laktobasil sayısı ve maya seviyesi kefir kullanımı ile birlikte gruplar arasında önemli farklılık ($P<0.001$) olmuştur. Yararlı bakteri sayısının artması ve zararlı bakterilerin azalması ile bağırsak mikroflorasında kefirin olumlu etkilerinin görüldüğü ve bu nedenle kefirin hayvanların sağlığı üzerinde olumlu etkisi olduğu sonucuna varıldı.

Anahtar sözcükler: *: Kefir, Kan parametreleri, Bağırsak mikroflora, Rat*

How to cite this article?

Ozsoy B, Cantekin Z, Yalcin S, Bayraktar HS: Effects of kefir on blood parameters and intestinal microflora in rats: An experimental study. *Kafkas Univ Vet Fak Derg,* 27 (1): 111-115, 2021.
DOI: 10.9775/kvfd.2020.24855

(*) **Corresponding Author**

Tel: +90 256 218 2000
E-mail: bulent58@gmail.com (B. Özsoy)

Introduction

Various feeds containing live microorganisms (probiotics) are now being used extensively and many researches are being carried out on their effects. Especially the effects of such food items on digestive system functions are being supported with new knowledge day by day. In the field of animal nutrition, new feed additives and performance enhancing substances are increasing progressively. These products are generally classified and named as prebiotics and probiotics. Kefir is one of Turkish traditional fermented dairy products, obtained by fermentation of ethyl alcohol and lactic acid using kefir extracts. Kefir is widely known in the Caucasus and has been produced by local people since ancient times. Since the 19th century, it has begun to be produced in many parts of the world. Kefir has a sharp acid taste and contains lactic acid, oxalic acid, little alcohol and lactic acid bacteria and some aromatic compounds as acetaldehyde and acetone that formed by yeasts [1-3]. Yeast flour is the main element which gives the unique taste of kefir. Since kefir is made from milk, it contains all the nutrients such as fat, lactose, minerals and vitamins in the milk. Even during the formation of certain vitamins, protein and lactose partial disintegration, kefir feed value is increasing. The microorganisms found in kefir composition enable this product to be digested easily so that the absorption of nutrients by the body is increasing. Kefir granules contain some microorganisms such as; lactobacilli, lactococci, leuconostocs, acetobacteria, and fungi (Kluyveromycesmarxianus, Torulasporadelbrueckii, *Saccharomyces cerevisiae*, Candida kefir [4,5]. The antioxidant, antifungal [6], antibacterial, antitumoral, immunological [7-9], triglyceride [10], and cholesterol lowering [11] and anti-apoptotic [12] effects of kefir were reported previously. Kefir microbiological composition which effects positively on human health has been reported [13-15]. By taking advantage of these properties of kefir, we aimed to examine its effects on intestinal microflora and some blood plasma parameters in rats.

Material and Methods

Ethical Approval

This study was conformed according to Hatay Mustafa Kemal University Animal Experiments Local Ethics Committee Presidency instructions and approved with consensus at the meeting (30/01/2014, 01/5).

Animal Sampling

A total of 24 female rats were used in this study. Rats were randomly allocated into one control group and three treatment groups each containing 6 rats. During 35 days of experimental period, rats were fed with a commercial diet having 23% crude protein and 2800 kcal/kg metabolizable energy. Feed in pellet form and water were provided *ad libitum*. Kefir was given at the levels of 10 mL/kg, 20 mL/kg and 30 mL/kg with oral gavage per day to the first, second and third treatment groups, respectively. Kefir was not given to the control group.

Detection of Yeast and Lactobacilli Count of Kefir

For counting of Yeast and Lactobacilli in kefir, a series of 10-fold dilutions were made by using sterile saline (FTS). For this purpose, four MRS Agar (de man, rogosa and sharpe) and four Sabouraud Dextrose Agar were inoculated with 100 µL from each dilution. The agar plates were incubated at 30°C for 72 h in aerobic conditions. After incubation, the cultures for each dilution were counted from the media and the averages were taken and the number of these microorganisms in kefir was determined [16]. The number of yeast was found to be $1.65x10^7$ and the number of lactobacilli was found to be $4x\ 10^8$ in kefir.

Determination of Microbial Flora and pH in Bowels

At the end of the 35-day study period, all animals were killed by decapitation under anesthesia. After the intestinal contents were homogenized, pH was measured (Orion Star Benchtop pH meter).

At the end of the process, the intestinal contents of 6 animals from each group were collected under sterile conditions and transferred into 50 mL sterile plastic tubes. Samples were diluted one-fold in FTS containing 0.9% $NaCl_2$, followed by serial 10-fold dilutions (log10) in FTS. Cultivated Plate Count Agar was incubated for 48 h at 30°C for Total Mesophilic Aerobic Microorganism (TMAM), MacConkey Agar for 24 h at 37°C for coliform bacteria count, and Sabouraud Dextrose Agar for 72 h at 30°C for yeast in aerobic conditions, MRS Agar for 72 h at 30°C in microaerophilic conditions for Lactobacilli. Four agar plates were used for each dilution. After the incubation the average was obtained by counting. The mean numbers for all dilution steps were then determined and the number of microorganisms in each sample was recorded [16].

Blood Analysis

At the end of the process, blood samples were collected from 6 animals in each group (total 24 rats) and transferred to EDTA tubes and centrifuged at 3000 rpm for 5 min. Plasma were then kept in deep freezing (-20°C). Total protein, albumin, uric acid, total cholesterol, triglyceride, alkaline phosphatase (ALP), serum glutamic oxaloacetic transaminase (SGOT), serum glutamic pyruvic transaminase (SGPT) and phosphorus analyzes were then performed with an autoanalyzer.

Statistical Analysis

Statistical analyses were done using SPSS programme (SPSS Inc., Chicago, IL, USA). Data for body weight, blood parameters, intestinal pH and microflora were analysed

as a completely randomized block design, with 4 dietary treatments and 6 samples using The effects of graded levels of kefir on these variables were analysed using polynomial contrasts. The significance of mean differences between groups were tested by Tukey. Level of significance was taken as $P<0.05$ [17].

Results

The number of yeasts was found to be $1.65x10^7$ and the number of lactobacilli was found to be $4x\ 10^8$ in kefir. The final body weight was found 322.83, 359.17, 338.00, 361.17 g in the control and experimental groups, respectively. There were no differences in the initial and final body weights among the groups as shown in *Table 1*. There were no statistical differences in total protein, albumin, uric acid, SGPT, SGOT, ALP and phosphorus among the groups. Triglyceride levels in the blood samples taken at the end of the trial were found to be 80.07, 79.90, 61.18, 61.52 mg/dL in the control and experimental groups, respectively. Blood cholesterol values were found to be 49.92, 46.62, 41.65, 38.72 mg/dL respectively. A linear decrease ($P<0.05$) was observed in blood plasma triglyceride and cholesterol levels with increasing kefir levels *(Table 2)*.

There were no differences among the groups in the pH values of intestinal contents *(Table 3)*. In the intestinal microflora, the number of Enterobacteria was the lowest in group 3 ($0.18x10^7$) and the highest in the control group

Table 1. *Effects of kefir on body weight of rats*

Parameters	Groups				Pooled SEM	Significance		
	Control (n=6)	Group 1 (n=6)	Group 2 (n=6)	Group 3 (n=6)		Linear	Quadratic	Cubic
Initial BW (g)	320.50	349.83	330.50	355.17	6.434	0.139	0.851	0.107
Final BW (g)	322.83	359.17	338.00	361.17	6.590	0.102	0.596	0.077

No significant differences among groups

Table 2. *Effects of kefir on blood plasma parameters in rats*

Parameters	Groups				Pooled SEM	Significance		
	Control (n=6)	Group 1 (n=6)	Group 2 (n=6)	Group 3 (n=6)		Linear	Quadratic	Cubic
Total protein mg/dL	48.80	48.33	49.68	49.08	0.430	0.589	0.941	0.358
Albumin mg/dL	30.22	29.22	28.13	28.80	0.375	0.118	0.267	0.581
Uric acid mg/dL	1.92	1.68	0.88	0.90	0.256	0.105	0.807	0.554
Triglyceride mg/dL	80.07^a	79.90^a	61.18^b	61.52^b	2.846	0.001	0.956	0.074
Cholesterol mg/dL	49.92^a	46.62^a	41.65^b	38.72^b	1.867	0.023	0.959	0.816
SGPT U/L	24.52	27.22	26.78	24.72	0.699	0.979	0.104	0.813
SGOT U/L	61.37	62.98	63.60	65.85	2.121	0.492	0.944	0.897
Alkaline phosphatase U/L	9.95	8.88	9.08	8.87	0.305	0.287	0.498	0.554
Phosphorus mg/dL	3.96	3.92	3.70	3.70	0.534	0.337	0.925	0.698

^{a,b} Means within a row followed by the different superscripts differ significantly ($P<0.05$)

Table 3. *Effects of kefir on intestinal pH and microflora (log10 cfu/g) in rats*

Parameters	Groups				Pooled SEM	Significance		
	Control (n=6)	Group 1 (n=6)	Group 2 (n=6)	Group 3 (n=6)		Linear	Quadratic	Cubic
pH	6.61	6.76	6.69	6.50	0.050	0.371	0.083	0.816
TMAM	57.40	57.20	63.60	57.60	1.428	0.585	0.317	0.150
Enterobacteria	3.16^a	2.40^{ab}	1.96^b	0.18^c	0.274	<0.001	0.050	0.142
Coliform	2.64^a	2.58^a	2.36^a	0.48^b	0.240	<0.001	0.004	0.234
Lactobacilli	1.66^c	2.92^b	3.20^b	7.52^a	0.513	<0.001	<0.001	<0.001
Yeast	0.17^b	0.20^b	0.28^a	0.35^a	0.019	<0.001	0.280	0.505

^{a,b,c} Means within a row followed by the different superscripts differ significantly ($P<0.05$)

($3.16x10^7$) and these findings were statistically significant ($P<0.001$) as shown in *Table 3*. Similarly, the number of coliform bacteria was found to be the lowest in group 3 ($0.48x10^7$). *Lactobacillus* counts were $1.66\ x10^7$ in the control group, $2.92\ x10^7$ in the first group, 3.22 in the second group and $7.52\ x10^7$ in the third group ($P<0.001$). The intestinal yeast level was the highest in the 3rd group ($0.35x10^7$). In summary; as the amount of kefir increased, there was a linear decrease in Enterobacteria, Coliform count while an increase in Lactobacilli and yeast counts was determined.

Discussion

In this study, effects of kefir were investigated on rats. There were no differences in body weights among the control and treatment groups. Sarı et al.[18] reported that the body weights of mice consumed the probiotic, kefir, kımız and yogurt were higher than that of control group. Karademir and Ünal [19] also concluded that there was a concordance between body weight gain and the amount of kefir in drinking water. Carnavelli et al.[20] fed sea bass with *Lactobacillius delbrueckii,* which they had isolated from the kefir, for 70 days and found that body weights of sea bass were increased with the consumption of *Lactobacillius delbrueckii*. Karademir et al.[21] have added kefir to laying hens' drinking water (0, 5, 7.5 and 10 mL/L) and they showed that kefir had a positive effect on egg shell thickness in the first period but it had no effects on other performance parameters.

In the present study there were no significant differences among the groups in total protein, albumin, uric acids, cholesterol, SGPT, SGOT, alkaline phosphatase and phosphorus in blood plasma. The triglyceride levels in groups treated 20 mL/kg and 30 mL/kg kefir daily were significantly lower than those of control group and the first group ($P<0.05$). Cholesterol and triglyceride levels were decreased linearly with increasing dose of kefir ($P<0.05$), but no significant differences in cholesterol level were observed among groups. The cholesterol levels in the groups treated 20 mL/kg and 30 mL/kg kefir daily were 16.6% and 22.4% lower than that of control group, respectively ($P>0.05$). The uric acid levels in the groups treated 20 mL/kg and 30 mL/kg kefir daily were 54.2% and 53.1% lower than that of control group, respectively ($P>0.05$). Adipose tissue produces and secretes uric acid through xanthine oxidoreductase and that its production is enhanced in obesity. Uric acid is a risk factor for cardiovascular diseases. Xanthine oxidase, is one of the enzymatic forms of xanthine oxidoreductase, induces oxidative stress in the manufacture of uric acid production. Thus, inhibition of xanthine oxidase suppresses the oxidative stress of uric acid that is related to cardiovascular diseases, obesity and insulin resistance [22,23]. The dose dependent significant decrease of total cholesterol level and triglyceride value by kefir administration was also reported previously [24,25]. However, Rattray and Connell [26], reported that plasma triglycerides were not affected by kefir consumption. Also Ozsoy [27] emphasized that kefir had positive effects on fatty liver in rats. Some researchers [28-30] reported that cholesterol lowering effect of kefir could be attributed to the deconjugation of bile acids by *Lactobacillus* spp.

In the present study, no differences were observed among groups in total mesophilic aerobic microorganism. But main differences were found in Lactobacili, yeast, Enterobacteiceae and total coliform counts. While Lactobacilli and yeast count increased, Enterobacteiceae and total coliform decreased with kefir consumption. Similarly, Yaman et al.[31] reported that no differences in total aerobic mesophilic bacteria, a significant ($P<0.05$) decrease of the coliform and Enterobacteriaceae population and significant increase in Lactobacilli count in the faeces of goslings supplemented with 0.2% and 0.5% kefir to drinking water. Zheng et al.[32] also reported that fecal lactobacilli counts were significantly ($P<0.05$) higher in rats fed by probiotic bacteria from tibetian kefir than in the control group. But, the amount of coliform organisms in the rat faeces was significantly decreased at day 28. After 28 days of administration, the amount of coliform organisms were remained stable until the end of 42 days. And they suggested that these strains may be used in the future as probiotic starter cultures for manufacturing novel fermented foods. Likewise, Yaman et al.[31] reported that *Lactobacillus* populations were significantly enhanced in geese receiving the highest dose of kefir in drinking water (0.5%) when compared with the controls. Colony forming units of *Enterobacteriaceae,* often associated with intestinal disease, were significantly lowered in the group with 0.5% kefir supplementation, indicating a Lactobacilli - *Enterobacteriaceae* antagonism, In the present study, a significant ($P<0.05$) decrease of the coliform population was observed. Wang et al.[24] reported that kefir administration at high cholesterol diets of rats did not affect intestinal pH parameters as like as our study. The differences in literatures may be due to the kefir usage in experiments, diets and animals.

As a result, the use of kefir in rats did not lead to any health problems. The blood level of triglyceride has been reduced considerably, and the number of beneficial bacteria in the intestine has been increased, while the number of harmful bacteria has been decreased. Since the most beneficial result was seen in the third trial group given 30 mL/kg kefir daily, consumption at these doses may be recommended.

Acknowledgement

This work was supported by Hatay Mustafa Kemal University Scientific Research Projects Coordinator (Grand number: 2015/13321) Turkey. This manuscript was presented in the congress indicated below and was published in abstract form in the proceedings of the congress *"32. World Veterinary Congress,* 13-17 September 2015, Istanbul".

Declaration of Interest Statement

We declare that we have no conflict of interest.

Authors Contributions

BO designed the experiments. BO, ZC, SY and HB performed the experiments and wrote the paper and analysed the data. BO provided the kefir.

References

1. Zubillaga M, Weill R, Postaire E, Goldman C, Caro R, Boccio J: Effect of probiotics and functional foods and their use in different diseases. *Nutr Res*, 21, 569-579, 2001. DOI: 10.1016/S0271-5317(01)00281-0

2. Kim DH, Kang IB, Jeong D, Kim H, Kim HS, Lee SK, Song KY, Seo KH: Development of rapid and highly specific TaqMan probe-based real-time PCR assay for the identification and enumeration of *Lactobacillus kefiri* in kefir milk. *Int Dairy J*, 61, 18-21, 2016. DOI: 10.1016/j.idairyj.2016.03.007

3. Kim DH, Jeong D, Song KY, Seo KH: Comparison of traditional and backslopping methods for kefir fermentation based on physicochemical and microbiological characteristics. *LWT*, 97, 503-507, 2018. DOI: 10.1016/j.lwt.2018.07.023

4. Kubo M, Odani T, Nakamura S, Tokumaru S, Matsuda H: Pharmacogical study on kefir a fermented milk product in Caucausus. I. On antitumor activity (1). *Yakugaku Zasshi*, 112, 489-495, 1992. DOI: 10.1248/yakushi1947.112.7_489

5. Neve H: Analysis of kefir grain starter cultures by scanning electron microscopy. *Milchwissenschaft*, 47, 275-278, 1992.

6. Hoolihan LK: Prophylactic and therapeutic use of probiotics: A review. *J Am Diet Assoc,* 101, 229-241, 2001. DOI: 10.1016/S0002-8223(01)00060-8

7. Garofalo C, Osimani A, Milanovic V, Aquilanti L, De Filippis F, Stellato G, Di Mauro S, Turchetti B, Buzzini P, Ercolini D, Clementi F: Bacteria and yeast microbiota in milk kefir grains from different Italian regions. *Food Microbiol*, 49, 123-133, 2015. DOI: 10.1016/j.fm.2015.01.017

8. Kim DH, Jeong D, Kang IB, Kim H, Seo KH: Development of a rapid and reliable TaqMan probe-based real-time PCR assay for the detection and enumeration of the multifaceted yeast *Kluyveromyces marxianus* in dairy products. *LWT*, 87, 163-168, 2018. DOI: 10.1016/j.lwt.2017.08.065

9. Eliş Yıldız S, Yiğit F, Duman Aydin B, Karadağ Sarı E, Deprem T, Koral Taşçı S: Effects of kefir, koumiss, milk and yoghurt administration on distribution of plasma cells and mast cells in mice spleen. *Kafkas Univ Vet Fak Derg*, 21, 195-201, 2015. DOI: 10.9775/kvfd.2014.12015

10. Mert H, Yılmaz H, Irak K, Yıldırım S, Mert N: Investigation of the protective effect of kefir against isoproterenol induced myocardial infarction in rats. *Korean J Food Sci Anim Resour*, 38, 259-272, 2018. DOI: 10.5851/kosfa.2018.38.2.259

11. St-Onge MP, Farnworth ER, Jones PJH: Consumption of fermented and nonfermented dairy products: Effects on cholesterol concentrations and metabolism. *Am J Clin Nutr,* 71, 674-681, 2000. DOI: 10.1093/ajcn/71.3.674

12. Matsuu M, Shichijo K, Okaichi K, Wen CY, Fukuda E, Nakashima M, Nakayama T, Shirahata S, Tokumaru S, Sekine I: The protective effect of fermented milk kefir on radiation-induced apoptosis in colonic crypt cells of rats. *J Radiat Res*, 44, 111-115, 2003. DOI: 10.1269/jrr.44.111

13. Nurliyani, Harmayani E, Sunarti: Antidiabetic potential of kefir combination from goat milk and soy milk in rats induced with streptozotocin-nicotinamide. *Korean J Food Sci Anim Resour*, 35, 847-858, 2015. DOI: 10.5851/kosfa.2015.35.6.847

14. Rosa DD, Dias MMS, Grzeskowiak LM, Reis SA, Conceiçao LL, Maria do Carmo GP: Milk kefir: Nutritional, microbiological and health benefits. *Nutr Res Rev*, 30, 82-96, 2017. DOI: 10.1017/S0954422416000275

15. Tung YT, Chen HL, Wu HS, Ho MH, Chong KY, Chen CM: Kefir peptides prevent hyperlipidemia and obesity in high-fat-diet-induced obese rats via lipid metabolism modulation. *Mol Nutr Food Res*, 62:1700505, 2018. DOI: 10.1002/mnfr.201700505

16. Arda M, Minbay A, Leloğlu N, Aydın N, Kahraman M, Akay Ö, Ilgaz A, İzgür M, Diker KS: Özel Mikrobiyoloji. 26, Medisan Yayınları, Ankara, 1997.

17. Dawson B, Trapp RG: Basic and Clinical Biostatistics, 3rd ed., 161-182, Lange Medical Books/McGraw-Hill Medical Publishing Division, New York, USA, 2001.

18. Sari EK, Bakir B, Aydin BD, Sözmen M: The effects of kefir, koumiss, yogurt and commercial probiotic formulations on PPARα and PPAR-β/δ expressions in mouse kidney. *Biotech Histochem,* 89, 287-295, 2014. DOI: 10.3109/10520295.2013.844274

19. Karademir G, Ünal Y: Broilerde kefirin probiyotik amaçla kullanılması. *Lalahan Hay Araşt Enst Derg,* 49, 47-54, 2008.

20. Carnevali O, Vivo L, Sulpizio R, Gioacchini G, Olivotto I, Silvi S, Cresci A: Growth improvement by probiotic in European sea bass juveniles *(Dicentrarchus labrax),* with particular attention to IGF-1, myostatin and cortisol gene expression. *Aquaculture,* 258, 430-438, 2006. DOI: 10.1016/j.aquaculture.2006.04.025

21. Karademir G, Yöruk MA, Tunç MA, Çelebi D: Yumurtacı tavuklarda kefirin performans ve yumurta kalitesine etkisi. *Atatürk Üniv Vet Bil Derg,* 7, 177-184, 2012.

22. Moriya C, Satoh H: Teneligliptin decreases uric acid levels by reducing xanthine dehydrogenase expression in white adipose tissue of male wistar rats. *J Diabetes Res,* 2016:3201534, 2016. DOI: 10.1155/2016/3201534

23. Tsushima Y, Nishizawa H, Tochino Y, Nakatsuji H, Sekimoto R, Nagao H, Shirakura T, Kato K, Imaizumi K, Takahashi H, Tamura M, Maeda N, Funahashi T, Shimomura L: Uric acid secretion from adipose tissue and its increase in obesity. *J Biol Chem,* 288, 27138-27149, 2013. DOI: 10.1074/jbc.M113.485094

24. Wang Y, Xu NV, Xi A, Ahmed Z, Zhang B, Bai X: Effects of *Lactobacillus plantarum* MA_2 isolated from Tibet kefir on lipid metabolism and intestinal microflora of rats fed on high-cholesterol diet. *Appl Microbiol Biotechnol,* 84, 341-347, 2009. *DOI:* 10.1007/s00253-009-2012-x

25. Özsoy B, Küçükgül A, Özsoy ŞY, Yumuşak N: Investigation the protective effects of kefir in experimental diabetes mellitus and nonalcoholic liver fattened rats. *Harran Üniv Vet Fak Derg*, 6, 142-146, 2017.

26. Rattray FP, Connell MJ: Fermented milks kefir. **In,** Fukay JW (Ed): Encyclopedia of Dairy Sciences. 2nd ed., 518-524, Academic Press San Diego, U.S.A., 2011.

27. Ozsoy SY: The protective effect of kefir on carbon tetrachloride-induced histopathological changes in the livers of rats. *Kafkas Univ Vet Fak Derg* 22, 403-408, 2016. DOI: 10.9775/kvfd.2015.14825

28. Begley M, Hill C, Gahan CGM: Bile salt hydrolase activity in probiotics. *Appl Environ Microbiol,* 72, 1729-1738, 2006. DOI: 10.1128/AEM.72.3.1729-1738.2006

29. Brashears MM, Gilliland SE, Buck LM: Bile salt deconjugation and cholesterol removal from media by *Lactobacillus casei. J Dairy Sci,* 81, 2103-2110, 1998. DOI: 10.3168/jds.S0022-0302(98)75785-6

30. Tamai Y, Yoshimitsu N, Watanabe Y, Kuwabara Y, Nagai S: Effects of milk fermented by culturing with varius lactic acid bacteria and a yeast on serum cholesterol level in rats. *J Ferment Bioeng*, 81, 181-182, 1996. DOI: 10.1016/0922-338X(96)87601-X

31. Yaman H, Ulukanli Z, Elmali M, Unal Y: The effect of a fermented probiotic, the kefir, on intestinal flora of poultry domesticated geese *(Anser anser). Revue Méd Vét*, 157, 379-386, 2006.

32. Zheng Y, Lu Y, Wang J, Yang L, Pan C, Huang Y: Probiotic properties of *Lactobacillus* strains ısolated from Tibetan kefir grains. *Plos One*, 8:e69868 2013. DOI: 10.1371/journal.pone.0069868