

# NUTRITION



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## Research Article Effects of Dietary Prebiotics (SafMannan) and Local Iraqi Prebiotic on Some Blood Biochemical Parameters of Broiler Chickens

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### Abstract

**Background and Objective:** This study was undertaken to investigate serum biochemical constituents of broiler chickens after incorporating a prebiotic (SafMannan, from Plileo Co.) and a local Iraqi prebiotic as feed additives. **Methodology:** A total of 195 straight run day-old broiler chicks (Ross 308) were randomly assigned to five experimental groups with 39 birds/group. Each group was subdivided into 3 replicates (13 birds/per replicate). The first group was fed a basal diet as a control group. The second and third groups were fed the basal diet supplemented with the SafMannan prebiotics at the level of  $250 \text{ g t}^{-1}$  (Saf 250) and  $500 \text{ g t}^{-1}$  (Saf 500), respectively. Meanwhile, the fourth and fifth groups were fed the basal diet supplemented with the local Iraqi prebiotics at the level of  $250 \text{ g t}^{-1}$  (IQP 250) and  $500 \text{ g t}^{-1}$  (IQP 500), respectively. **Results:** The results revealed a significant ( $p \le 0.05$ ) effect of SafMannan (Saf) and local Iraqi prebiotics (IQP) on the decrease in serum glucose, cholesterol, triglyceride concentration and aspartate transaminase (AST) enzyme activity in all dietary groups compared to the control. The group of Saf 250 and IQP 250 showed higher ( $p \le 0.05$ ) total protein and high-density lipoprotein cholesterol (HDL-c) levels. **Conclusion:** Supplementation of broiler chickens' diets with 250 and 500 g t<sup>-1</sup> of SafMannan prebiotics and Iraqi prebiotics led to improve serum lipid profiles and serum protein parameters. SafMannan prebiotics performed best by influencing serum blood indices compared to local Iraqi prebiotics.

Key words: Broiler, prebiotic, SafMannan, lipid profile, liver enzyme, serum protein

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Competing Interest: The author has declared that no competing interest exists.

Data Availability: All relevant data are within the paper and its supporting information files.

#### INTRODUCTION

Currently, administration of antimicrobials to animals leads to enhanced performance of animals and to the suppression of pathogenic bacterial growth. However, there are increasingly bans on the use of antimicrobials and antibiotics. Researchers in the field of poultry nutrition have focused on studying some non-dietary additives that have had significant impacts in supporting the physiological status and productivity of birds. These include using types of probiotics, prebiotics and synbiotics in poultry diets. Manna-oligosaccharides (MOS) and fructo-oligosaccharides (FOS) have both been investigated for prebiotic activities in broilers<sup>1</sup>. Safmannan is a source of mannan-oligosaccharide and β-glucans derived from the yeast cell wall (YCW) of Saccharomyces cerevisiae. Many yeast companies currently manufacture YCW. Previous studies showed the positive effect of YCW on starter broiler<sup>2-5</sup> and laying hen performance<sup>6</sup>. They showed that a dose of 250 mg kg<sup>-1</sup> of SafMannan led to optimum beneficial effects. Supplementation with MOS to laying hen diets led to improved performance, increased lactobacillus and decreased salmonella and enhanced nutrient digestibility<sup>7</sup>. Gao et al.<sup>8</sup> showed that feeding broilers the basal diet supplemented with YCW 2.5 g kg<sup>-1</sup> from hatching time to 24 days led to enhanced daily gain and feed conversion ratio. Gao et al.8 also found increased villi height/crypt width ratio of the ileum on day 21 and the jejunum and duodenum on day 42 of age. Baurhoo et al.9 suggested that the supplementation of Bio-Mos<sup>®</sup> in the diets increased intestinal villus height and improved colonization of beneficial bacteria in broilers. Supplementation of Bio-Mos® altered intestinal microbiota of broilers without affecting performance<sup>10</sup>. Additionally, dietary inclusion of yeast cell wall containing manna oligosaccharides (YCW-MOS) led to improved body weight with reduced feed efficiency<sup>11</sup>. Sahin et al.<sup>12</sup> reported that the inclusion of Synbiotic to broiler diets did not have any influence on total serum protein, albumin and total cholesterol levels. The addition of 0.3 and/or 0.4% synbiotic has a positive effect on guails by improving performance and enhancing some serum lipids and protein parameters. Prebiotics have a positive effect on cholesterol reduction. Owing to the non-digestible nature of prebiotics, fermentation occurs in the intestine, leading to increased activity and growth of one or more probiotics<sup>13-15</sup>. Prebiotics eliminate cholesterol in the intestine through reducing lipid absorption by the action of bile salt hydrolase enzyme<sup>16</sup>. Bile salt hydrolase enzyme is an enzyme that is responsible for bile salt deconjugation of the cholesterol from bile, thus decreasing the emulsification of lipids<sup>17</sup>. The current

study was designed to evaluate the effects of dietary supplementation of a prebiotic (SafMannan) and a local Iraqi prebiotic on some biochemical constituents of blood serum of broiler chickens.

#### **MATERIALS AND METHODS**

Animal husbandry and treatments: This study was conducted at the poultry farm of the Animal Production Department, College of Agriculture, AL-Qasim Green University, from 22 October to 25 November, 2017. A total of 195 straight run one-day-old broiler chicks (Ross 308) were assigned randomly to five experimental groups in cages (39 birds/group). Each group contained three replicates (13 birds/replicate). The first group was fed a basal diet and served as a control group. The second and third groups were fed the basal diet supplemented with the prebiotics (SafMannan) at the level of 250 and 500 g t<sup>-1</sup>, respectively. Meanwhile, the fourth and fifth groups were fed the basal diet supplemented with the local Iragi prebiotics at the level of 250 and 500 g t<sup>-1</sup>, respectively. Basal diets were formulated to meet or exceed the nutrient requirements of the broilers<sup>18</sup>. The formulation of the diet is shown in Table 1. The birds were fed a starter diet for 21 days, followed by a finisher diet from days 22-35. All birds received diet ad libitum during the 35-day period. SafMannan® was bought from the Phileo Lesaffre Animal Care Company. It is a yeast cell wall from Saccharomyces cerevisiae and is rich in β-glucans and mannan-oligosaccharides. The Iraqi local prebiotic used in this experiment was from yeast Saccharomyces cerevisiae (non-starch oligosaccharide) after destruction of the cell wall of the yeast<sup>19</sup>. No drug therapy was received during the experiment. All chicks received vaccines on the day of hatching in the hatchery. General flock condition, ration, water, temperature, mortality and unexpected events were observed daily.

**Biochemical parameters evaluation:** Three chicks from each replicate were randomly selected at the end of the experiment to determine blood serum parameters. Blood samples were collected separately in non-heparinized tubes for biochemical assays. All tubes were centrifuged (3000 rpm) for 15 min at 25°C to obtain serum. Serum samples were stored at -20°C until analyzed for total protein and albumin by a colorimetric method using commercial kits (Bio lab AS, France). The serum globulin was calculated as the difference between serum total protein and albumin. Blood serum cholesterol, triglycerides and glucose concentrations were determined according to the methods of Zangana and Naji<sup>19</sup> using commercial kits (Biolabo

Ingredients (%)	Starter diet (1-21 days)			Finisher diet (22-35 days )				
	Control	(Pre 0.25%)	(Pre 0.5%)	Control	(Pre 0.25%)	(Pre 0.5%)		
Yellow corn	30.00	30.000	30.00	30.00	30.000	30.00		
Wheat	27.50	27.475	27.45	35.50	35.475	35.45		
Soybean meal (44%)	31.00	31.000	31.00	23.00	23.000	23.00		
Vegetable oil	3.00	3.000	3.00	3.00	3.000	3.00		
Protein concentrate (44%) <sup>1</sup>	7.00	7.000	7.00	7.00	7.000	7.00		
Limestone	1.00	1.000	1.00	1.00	1.000	1.00		
Vitamin premix <sup>2</sup>	2.50	2.500	2.50	2.50	2.500	2.50		
Prebiotic Saf <sup>3</sup> or IQP	-	0.025	0.05	-	0.025	0.05		
Sodium chloride	0.25	0.250	0.25	0.25	0.250	0.25		
Total	100.00	100.000	100.00	100.00	100.000	100.00		
Metabolizable energy (kcal kg <sup>-1</sup> )	3032.00	3032.000	3032.00	3157.00	3157.000	3157.00		
Crude protein (%)	24.32	24.320	24.32	23.00	23.000	23.00		
Calorie: protein ratio	124.67	124.670	124.67	137.20	137.200	137.20		
Ether extract (%)	5.90	5.900	5.90	6.17	6.170	6.17		
Crude fiber (%)	3.68	3.680	3.68	3.33	3.330	3.33		
Calcium (%)	1.10	1.100	1.10	1.10	1.100	1.10		
Phosphorus available (%)	0.48	0.480	0.48	0.51	0.510	0.51		
Lysine (%)	1.22	1.220	1.22	1.10	1.100	1.10		
Methionine (%)	0.42	0.420	0.42	0.38	0.380	0.38		

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Table 1: Ingredients and nutrient composition of broiler starter and finisher diets

<sup>1</sup>Protein concentrate used from Al-Hayat company, Jordanian origin, to provide the following per kg of diet: Protein: 44%, ME: 2800 kcal kg<sup>-1</sup>, Fat: 12%, Ash: 25%, Calcium: 5%, Phosphorus: 2.9%, Methionine: 2.55%+cysteine, Lysine: 2.8%. <sup>2</sup>Each kilogram of feed (Provimi Jordan Co., Amman, Jordan) contained the following: Iron: 44 mg, Copper: 5 mg, Zinc: 75 mg, Manganese: 6 mg, Iodine: 1.306 mg, Selenium: 0.225 mg, Folic acid: 0.6 mg, Biotin: 100 g, Pantothenic acid: 10 mg, Niacin: 39.994 mg, Vitamin A (retinyl acetate): 12,500 IU, Vitamin D3: 2,500 IU, Vitamin E: 50 IU, Vitamin K3: 3.5 g, Vitamin B1: 1 g, Vitamin B2: 5.5 g, Vitamin B6: 2.5 g, Vitamin B1: 2.00 g. <sup>3</sup>Safmannan<sup>®</sup> from Phileo Lesaffre animal care company

AS, France). The concentration of high-density lipoprotein cholesterol in the serum was estimated by a previously published method of Tietz<sup>20</sup>. The activities of serum Aspartate aminotransferase (AST) and Alanine aminotransferase (ALT) were measured using diagnostic kits (QCA, Amposata, Spain). The AST/ALT ratio was calculated.

**Statistical analysis:** Data were analyzed using one way ANOVA by SPSS<sup>21</sup>. Difference between treatment means were assessed using the Least Significant Difference (LSD) test at 5% level of significance<sup>22</sup>.

#### RESULTS

The effect of Saf-Mannan prebiotic (SAF) and a local lraqi prebiotic (IQP) on some blood indices at the end of the experiment is shown in Table 2. A significant ( $p \le 0.05$ ) difference was recorded for serum attributes. Serum glucose, cholesterol, triglyceride concentrations and AST enzyme activity declined in all dietary groups compared with the control. However, the group given 250 g t<sup>-1</sup> SAF showed a higher significant level ( $p \le 0.05$ ) of total protein than the control. Globulin levels were similar (p > 0.05) in the treated groups compared with control group. Chicks that were fed 250 g t<sup>-1</sup> IQP had the highest albumin/globulin ratio compared with the 250 g t<sup>-1</sup> SAF and control groups. Serum

albumin (ALB) and high-density lipoprotein cholesterol (HDL-c) concentrations increased significantly ( $p \le 0.05$ ) when SAF and IQP were included in the broiler diets. The group given 500 g t<sup>-1</sup> SAF showed lower ALT activity significantly ( $p \le 0.05$ ) than the control. In addition, chicks that received the 250 g t<sup>-1</sup> SAF showed a lower value of the AST/ALT ratio compared to the control.

#### DISCUSSION

In the process of anabolism and proteolysis of protein, serum total protein was related to the reflection of dietary protein and immune response. The elevation of serum total protein values (TP) in treated groups with prebiotic may be attributed to the utilization of organic compounds of feed intake, especially protein. These results are in accordance with the previous studies<sup>23-27</sup>, which showed the same trend by feeding prebiotics<sup>28,29</sup> during feeding of synbiotics. The inclusion of 1% prebiotic inulin in the diet had no effect on TP, ALB, urea, creatinine, glucose and blood serum amylase in male *coturnix* quails<sup>30</sup>. A reduction in circulating glucose with supplemental SAF and IQP showed similar results as described by Al-Kassie et al.<sup>31</sup> who reported that the supplementation of probiotics (Aspergillus niger) and prebiotics (Taraxacum officinale) (10 g kg<sup>-1</sup>) to broiler chicks resulted in a significant decrease in glucose concentration. In another study<sup>32</sup>, the

Parameters	Dietary groups							
	Control (0 g t <sup><math>-1</math></sup> )	SAF (250 g t <sup>-1</sup> )	SAF (500 g t <sup>-1</sup> )	IQP (250 g t <sup>-1</sup> )	IQP (500 g t <sup>-1</sup> )			
Total protein (g dL <sup>-1</sup> )	3.54±0.32 <sup>b</sup>	4.22±0.15ª	4.05±0.12 <sup>ab</sup>	3.86±0.14 <sup>ab</sup>	3.79±0.13 <sup>ab</sup>			
Albumin (g dL <sup>-1</sup> )	1.39±0.06°	1.94±0.29 <sup>b</sup>	2.52±0.02ª	2.50±0.17ª	2.35±0.05ªb			
Globulin (g dL <sup>-1</sup> )	2.15±0.26 <sup>ab</sup>	2.28±0.44ª	1.53±0.13 <sup>ab</sup>	1.36±0.14 <sup>b</sup>	1.44±0.18 <sup>b</sup>			
Albumin to globulin ratio	0.72±0.38°	1.00±0.40 <sup>bc</sup>	1.66±0.15 <sup>ab</sup>	1.88±0.20ª	1.68±0.24 <sup>ab</sup>			
Glucose (mg dL <sup>-1</sup> )	274.83±7.27ª	219.21±1.81°	233.22±2.59 <sup>bc</sup>	241.99±5.13 <sup>b</sup>	243.99±4.95 <sup>b</sup>			
Cholesterol (mg dL <sup>-1</sup> )	142.36±8.88ª	112.19±2.74 <sup>b</sup>	111.57±3.83 <sup>b</sup>	121.69±2.28 <sup>b</sup>	126.74±3.20 <sup>b</sup>			
Triglyceride (mg dL <sup>-1</sup> )	130.55±2.98ª	51.98±6.37 <sup>d</sup>	54.02±7.56 <sup>d</sup>	106.57±4.05 <sup>b</sup>	74.09±6.34°			
HDL-c (mg dL <sup>-1</sup> )	58.66±3.86°	90.06±5.95ª	91.86±5.20ª	74.05±2.74 <sup>b</sup>	74.78±1.61 <sup>b</sup>			
$AST(U L^{-1})$	107.50±9.69ª	81.39±6.31 <sup>bc</sup>	79.01±4.96°	92.20±6.42 <sup>b</sup>	91.70±5.74 <sup>b</sup>			
$ALT(U L^{-1})$	10.37±0.54ª	9.59±0.55ab	8.41±1.09 <sup>b</sup>	9.24±0.62 <sup>ab</sup>	$9.57 \pm 0.48^{ab}$			
AST/ALT ratio	10.34±0.49ª	8.49±0.73 <sup>b</sup>	$9.45 \pm 0.71^{ab}$	10.02±1.26ª	9.58±0.67 <sup>ab</sup>			

Table 2: Some serum blood characteristics at 35 days of	of age of broilers fed (SAF) and (IQP) (Means $\pm$ SE)
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<sup>a-d</sup>Means in the same row with no common superscript are significantly different ( $p \le 0.05$ ), HDL-c: High-density lipoprotein cholesterol, AST: Aspartate aminotransferase enzyme, ALT: Alanine aminotransferase enzyme

addition of 1.5% yeast prebiotic to a broiler diet decreased serum glucose. Abd-El-Rahman et al.26 found no change in glucose levels with the inclusion of Bactocell and Revitilyte-Plus as a probiotic in broiler chicken diets. The supplementation of SAF and IQP led to a decrease in serum cholesterol and triglyceride levels. This may be attributed to reduced absorption and/or synthesis of cholesterol in the gastrointestinal tract by probiotic supplementation<sup>33</sup>. Brashears et al.34 indicated that most of the cholesterol removed from broth by Lactobacillus acidophilus L. by means of the destabilization of cholesterol micelles and the co-precipitation of the cholesterol with the deconjugated bile salts at pH less than 6. In addition, the prebiotics may possess the property of reducing cholesterol in the blood by inhibition of its hepatic synthesis or by deconjugating the biliary salts<sup>29</sup>. Moreover, inhibition of 3HMG-CoA, which is an intermediate of mevalonate during the synthesis of cholesterol from acetyl-Co A by fermented milk products (Kefiran), has been suggested as the reason for the reduced level of cholesterol in serum<sup>35</sup>. The effects of probiotic, prebiotic and symbiotic on reducing total cholesterol levels were shown by various studies<sup>26,29,36-38</sup>. Meanwhile, the increase in HDL-c levels was shown by other studies<sup>29,39</sup>. Contrary to the results of this study, some previous studies have reported no effects on the TC, triglyceride and HDL-c levels<sup>11,40</sup>. Results of the present study showed that dietary supplementation of prebiotic SAF and IQP can lower serum lipids in broilers. Adding SAF and IQP not only reduced triglycerides but also led to decreases in cholesterol and an increase in serum HDL. These findings are in agreement with the study of Li et al.<sup>23</sup> who reported that adding the prebiotic chito-oligo saccharide led to decreased serum cholesterol and triglycerides. Prebiotics have a positive effect on cholesterol reduction. Because of the non-digestible nature of prebiotics, fermentation occurs in the intestine, leading to increased activity and growth of one or more probiotics<sup>13-15</sup>. Prebiotics eliminate cholesterol in the intestine through reducing lipid absorption by the action of the bile salt hydrolase enzyme<sup>16</sup>. Bile salt hydrolase enzyme is an enzyme that is responsible for bile salt deconjugating of the cholesterol from bile, thus decreasing emulsification of lipids<sup>17</sup>. Inclusion of SAF and IQP in broiler diets decreased ( $p \le 0.05$ ) AST levels. These results are similar to Aluwong et al.<sup>32</sup>, who recorded that supplementation of broiler feeds with 2.0% yeast decreased the activity of serum ALT. Tufan and Bolacali<sup>29</sup> found a significant reduction in AST levels in quails after 42 days of feeding 0.1% synbiotic. However, Abd-El-Rahman et al.,<sup>26</sup> showed a significant increase in AST and ALT enzyme activity in broilers that received prebiotics. In addition, no significant effect was reported for prebiotic, probiotic and synbiotic supplements on serum ALT and AST values of broiler chicks<sup>41</sup>.

#### CONCLUSION

It is concluded that supplementation of basal diets with prebiotics (Saf-Mannan) and local Iraqi prebiotics has a beneficial effect on the blood parameters of broiler chickens, with respect to decreasing serum cholesterol, triglyceride concentration and aspartate amino transferase enzyme activity and increasing HDL-c, total protein and albumen levels, which is important for maintaining the health of birds. According to the results of the current study, it appears that Saf-Mannan prebiotics are the best at influencing blood indices compared to local Iraqi prebiotics.

#### SIGNIFICANCE STATEMENT

This study discovers the possible effect of SafMannan and an Iraqi prebiotic that can be beneficial for estimating some blood biochemical parameters of broiler chicks (Ross 308). This study will help researchers to uncover the critical areas of the use of prebiotics as feed additives that many researchers have not been able to explore. This is a new theory about the non-digestible nature of prebiotics. Fermentation occurs in the intestine, leading to the elimination of cholesterol in the intestine through the reduction of lipid absorption by the action of bile salt hydrolase enzyme, which is responsible for bile salt deconjugating of the cholesterol from bile, thus decreasing emulsification of lipids.

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