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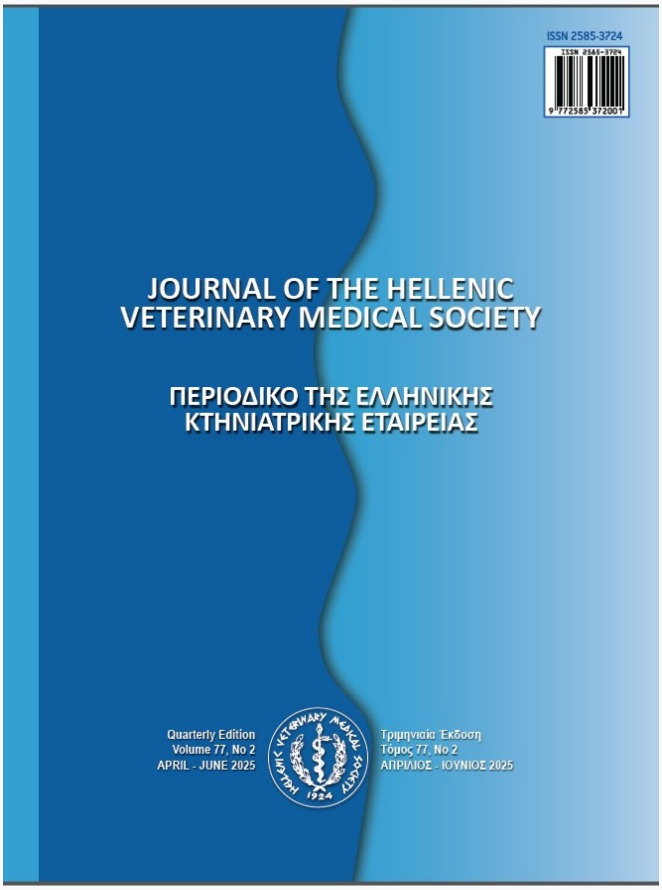

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Research article
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The effect of addition of powder jujube (*Ziziphus jujuba Mill.*) fruit at different rates into mixed diets on antioxidant and some blood parameters of broiler chickens*

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ABSTRACT: This study was conducted to determine the effects of addition of jujube (*Ziziphus jujuba Mill.*) fruit at different rates into the mixed feed on intestinal morphology, antioxidant and some blood parameters of broiler chickens. A total of 160 male broiler (initial body weights were determined as 209.25±2.17, 209.18±2.13, 209.18±1.74 and 209.20±2.58, respectively) chickens (Ross-308), including 40 in each group, were used in the study. The groups were divided into 4 sub-groups, each of which include 10 broiler chickens. The study groups were the control group (the group without jujube fruit), the H-0.5% group (the group with 0.5% jujube fruit), the H-1 group (the group with 1% jujube fruit), and the H-2 group (the group with 2% jujube fruit). At the end of the experiment (42nd day), the control group had the highest levels of blood glucose, triglycerides, total cholesterol, very low density lipoprotein cholesterol, alanine aminotransferase, aspartate aminotransferase, and uric acid, and the lowest level of creatine ($P<0.05$). While the lowest malondialdehyde level was detected in the H-2 group ($P<0.001$), the highest glutathione peroxidase ($P<0.01$) and catalase ($P<0.05$) levels were observed in the H-1 and H-2 groups. The control group also had the lowest values for villus height and crypt depth and the highest values for villus height/crypt depth ($P<0.001$). As a result, it was concluded that jujube fruit, which is mainly used for its antioxidant properties, can be used in poultry compound feeds especially at the level of 1% due to its positive effects on antioxidant and blood parameters and intestinal morphology.

Keyword: Antioxidant; blood parameters; broiler chicken; jujube fruit.

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INTRODUCTION

Proteins of animal origin have an important place in human nutrition. Since it is difficult to obtain essential amino acids, which must be taken from outside for a balanced diet, only from plant sources, the demand for animal sources is increasing day by day. Red and white meat particularly meet these needs. White meat is budget-friendly because it is produced faster and at a lower cost than red meat is. Chicken meat contains proteins and amino acids valuable to human health and is therefore one of the main sources of a balanced diet (Ambaw et al., 2021). Its high protein, vitamin and mineral content and low-fat content have made chicken meat useful for people of all ages (Franca et al., 2015). Poultry meat is a suitable source for reducing the risk of developing diseases related to the circulatory system, including cardiovascular diseases. Collagen produced from poultry is used to control hypertension (Franca et al., 2015, López et al., 2019). Chicken meat is has been widely used to prevent obesity (Astrup et al., 2014). Because poultry meat has high protein and low fat content, people consume this meat to effectively reduce obesity (Franca et al., 2015). Jujube is a plant that belongs to Rhamnaceae family and has more than 135 species (Liu et al., 2020). The most commonly cultivated species for their fruits are *Ziziphus jujuba* and *Ziziphus mauritiana*, and the fruits obtained from them are consumed fresh or dried (Gao et al., 2013; Wang et al., 2016). Jujube is also used in traditional medicine and as a food additive due to its rich nutritional content and bio-functional properties of its components (Xue et al., 2009; Choi et al., 2011). Bioactive substances such as vitamin C, flavanoids and polysaccharides contained in jujube fruit are thought to be natural antioxidants due to their free radical scavenging effects (Gao et al., 2003; Zhang et al., 2010). The antioxidant effect of jujube arises as a result of binding of phenolic compounds in its structure to free radicals, binding metal ions and inactivating lipoxygenase enzyme (Oguz, 2008).

The aim of this study is to identify the effects of adding jujube fruit powder, which has strong antioxidant properties, into broiler compound feeds at different levels on antioxidant and some blood and histological parameters.

MATERIALS AND METHODS

Research and Publication Ethics

The study was approved by Fırat University Animal Experiments Local Ethics Committee (FUHADYEK) (Date: 22.06.2021 - Protocol No: 2021/2633).

Location of the Study

The experiment was conducted in an environmentally controlled broiler house numbered TR230000360680 operating in Elazığ province. During the experiment, the animals were fed in the poultry house with the same ventilation, light intensity (20 lux) and other maintenance conditions. Broiler chickens were reared in ventilated poultry houses containing 16 compartments (10 broilers/m^2) and wood shavings were used as litter material. They were heated using electric radiators. In the first week, the temperature of these houses was kept between 32-35°C and gradually decreased to 22°C until the end of the experiment. They were illuminated for 24 hours with natural daylight during the day and fluorescent lamps at night.

Animal Material

In this study, 160 day-old male broiler chicks (ROSS 308) purchased from a private poultry farm were used as animal material.

Feed Material

The feeds used in the experiment were prepared by a feed factory. For this purpose, taking into account the requirements specified in NRC (1994) standards, compound feeds containing corn and soybean meal were prepared as isonitrogenous and isocaloric in 3 periods as starting (between days 0-21), growing (between days 22-35) and finishing (between days 36-42). Tables 1, 2, and 3 show the structure and nutrient composition of the compound feeds.

Jujube Added to Mixed Feed

Jujube fruit added to the mixed feed was obtained from a grower in Hatay province. Fully ripe fruits were collected in August, dried and then ground together with the seeds. Having been collected, dried and ground in season, jujube fruit was added to compound feeds as powder. Tables 5 and 6 show raw nutrient and chemical compositions of jujube fruit.

Experimental Design

The study was carried out on a total of 160 male chicks at 7 days of age. They were assigned to the groups as 40 animals in each. The chicks were randomly distributed to 4 experimental groups in accordance with the random plots experimental design and each group had 4 replicates. In this context, after the initial body weights of the chicks were measured,

Table 1. Ingredients and nutrient composition of experimental diets (0-21 days) (%)

	Jujube, %			
Feed Ingredients	**Control**	**0.5**	**1**	**2**
Maize	46.10	46.15	45.65	45.65
Soybean meal (%48 HP)	38.00	38.00	38.00	38.00
Wheat bran	6.55	6.00	6.00	5.00
Vegetable Oil	5.00	5.00	5.00	5.00
Dicalcium phosphate (DCP)	1.70	1.70	1.70	1.70
Ground limestone	1.15	1.15	1.15	1.15
Salt	0.35	0.35	0.35	0.35
DL- Methionine	0.35	0.35	0.35	0.35
L-Lysine hydrochloride	0.20	0.20	0.20	0.20
L-Threonine	0.10	0.10	0.10	0.10
Vitamin-Mineral mix*	0.50	0.50	0.50	0.50
Jujube fruit	0	0.50	1.00	2
Nutritional composition, (%)				
Dry matter	90.38	90.20	90.45	90.60
Crude protein	23.50	23.40	23.30	23.20
Crude fibre	3.65	3.75	3.78	3.56
Ether extract	4.92	5.15	5.08	4.98
Crude ash	5.44	5.66	5.55	5.38
Calcium**	0.97	0.97	0.97	0.97
Available Phosphorus**	0.49	0.49	0.49	0.49
Sodium**	0.17	0.17	0.17	0.17
Chlorine**	0.25	0.25	0.25	0.25
Methionine+Cystine**	1.10	1.10	1.10	1.10
Lysine**	1.44	1.44	1.44	1.44
Threonine**	0.97	0.97	0.97	0.97
Tryptophan**	0.33	0.33	0.33	0.33
ME, kcal/kg**	3044	3039	3022	3009

*: Per kg: Vitamin A, 12000 IU; Vitamin D3, 3000 IU; Vitamin E, 30 mg; Manganese, 80 mg; Iron, 60 mg; Zinc, 60 mg; Copper, 5 mg; Iodine, 1.5 mg; Cobalt, 0.3 mg; Selenium 0.15 mg **: Obtained by calculation.

average body weights of the groups were arranged in such a way that they were equal and a distribution was made with 10 chicks in each replicate and a total of 40 animals in each experimental group. Accordingly, the study groups were determined as the control group (the group without jujube fruit), the H-0.5% group (the group with 0.5% jujube fruit), the H-1 group (the group with 1% jujube fruit), and the H-2 group (the group with 2% jujube fruit). Initial body weights and percentage variances of chicks were determined as 209.25±2.17 (6.87%), 209.18±2.13 (6.75%), 209.18±1.74 (6.48%) and 209.20±2.58 (6.75%) in control, H-0.5%, H1% and H2% groups, respectively. The animals were fed water and feed ad libitum during the experiment.

Measurements and Analyses

Crude nutrient levels of compound feed were determined in the laboratories of Fırat University, Faculty of Veterinary Medicine, Department of Animal Nutrition and Nutritional Diseases. Dry matter, crude ash, crude protein, crude fat, and non-nitrogenous solids of compound feeds were analysed according to the analysis methods reported in AOAC (1980) and crude cellulose content was determined according to Crampton and Maynard (1983). The ration pro-

Table 2. Ingredients and nutrient composition of experimental diets (22-35 days) (%)

		Jujube, %		
Feed Ingredients	**Control**	**0.5**	**1**	**2**
Maize	49.35	49.15	49.15	49.15
Soybean meal (%48 HP)	33.00	33.50	33.50	34.00
Wheat bran	7.90	7.05	6.55	4.85
Vegetable Oil	5.70	5.80	5.80	6.00
Dicalcium phosphate (DCP)	1.55	1.55	1.55	1.55
Ground limestone	1.00	1.00	1.00	1.00
Salt	0.35	0.35	0.35	0.35
DL- Methionine	0.35	0.30	0.30	0.30
L-Lysine hydrochloride	0.20	0.20	0.20	0.20
L-Threonine	0.10	0.10	0.10	0.10
Vitamin-Mineral mix*	0.50	0.50	0.50	0.50
Jujube fruit	0	0.50	1.00	2
Nutritional composition, (%)				
Dry matter	90.30	89.90	89.54	89.68
Crude protein	21.62	21.65	21.60	21.50
Crude fibre	3.85	3.15	3.78	3.75
Ether extract	5.42	5.15	5.18	5.28
Crude ash	4.44	4.66	4.95	4.74
Calcium**	0.87	0.87	0.87	0.87
Available Phosphorus**	0.45	0.45	0.45	0.45
Sodium**	0.17	0.17	0.17	0.17
Chlorine**	0.25	0.25	0.25	0.25
Methionine+Cystine**	0.99	0.99	0.99	0.99
Lysine**	1.32	1.32	1.32	1.32
Threonine**	0.89	0.89	0.89	0.89
Tryptophan**	0.30	0.30	0.30	0.30
ME, kcal/kg**	3116	3115	3109	3118

*: Per kg: Vitamin A, 12000 IU; Vitamin D3, 3000 IU; Vitamin E, 30 mg; Manganese, 80 mg; Iron, 60 mg; Zinc, 60 mg; Copper, 5 mg; Iodine, 1.5 mg; Cobalt, 0.3 mg; Selenium 0.15 mg, **: Obtained by calculation.

gramme (Poultry_V5.05) prepared by Coşkun et al., (2019) was used to calculate the energy, mineral, and amino acid levels of the mixed feed. Crude nutrient analysis of the powdered additive obtained from jujube fruit was carried out by Makimtek Special Food Control Laboratory of the Ministry of Agriculture and Forestry of the Republic of Turkey.

At the end of the experiment, a total of 32 broiler chickens including two from each sub-group and eight from each group, were selected, and then they were starved for 10 hours and slaughtered using the neck-cutting technique. Blood samples were taken in gel centrifuge tubes from the broiler chickens during slaughtering and were centrifuged at 3500 rpm for 15 minutes to obtain serum samples. Serum samples were analysed for glucose, triglyceride, total cholesterol, VLDL (very low-density lipoprotein) cholesterol, alanine aminotransferase (ALT), aspartate aminotransferase (AST), uric acid and creatine using a biochemical analyser ADVIA 1800 in the Haematology Department of the Biochemistry Department of Fırat University Medical Faculty.

The determination of oxidative stress and antioxidant parameters in liver tissue obtained from 32 animals slaughtered at the end of the experiment was carried out in the laboratories of the Department of

Table 3. Ingredients and nutrient composition of experimental diets (36-42 days) (%)

		Jujube %		
Feed Ingredients	**Control**	**0.5**	**1**	**2**
Maize	53.50	53.50	53.50	52.00
Soybean meal (%48 HP)	29.50	29.50	30.00	30.40
Wheat bran	6.75	6.25	5.25	5.00
Vegetable Oil	6.45	6.45	6.45	6.80
Dicalcium phosphate (DCP)	1.35	1.35	1.35	1.35
Ground limestone	1.00	1.00	1.00	1.00
Salt	0.35	0.35	0.35	0.35
DL- Methionine	0.30	0.30	0.30	0.30
L-Lysine hydrochloride	0.20	0.20	0.20	0.20
L-Threonine	0.10	0.10	0.10	0.10
Vitamin-Mineral mix*	0.50	0.50	0.50	0.50
Jujube fruit	0	0.50	1.00	2
Nutritional composition (%)				
Dry matter	89.33	89.95	89.74	89.62
Crude protein	20.10	20.00	20.10	20.10
Crude fibre	3.65	3.75	3.88	3.95
Ether extract	5.70	5.55	5.48	5.68
Crude ash	3.99	4.16	3.95	3.74
Calcium**	0.81	0.81	0.81	0.81
Available Phosphorus**	0.41	0.41	0.41	0.41
Sodium**	0.17	0.17	0.17	0.17
Chlorine**	0.25	0.25	0.25	0.25
Methionine+Cystine**	0.95	0.95	0.95	0.95
Lysine**	1.21	1.21	1.21	1.21
Threonine**	0.83	0.83	0.83	0.83
Tryptophan**	0.27	0.27	0.27	0.27
ME, kcal/kg**	3220	3213	3213	3201

*: Per kg: Vitamin A, 12000 IU; Vitamin D3, 3000 IU; Vitamin E, 30 mg; Manganese, 80 mg; Iron, 60 mg; Zinc, 60 mg; Copper, 5 mg; Iodine, 1.5 mg; Cobalt, 0.3 mg; Selenium 0.15 mg, **: Obtained by calculation.

Physiology, Faculty of Veterinary Medicine, Fırat University. Lipid peroxide (MDA) in liver tissue was identified according to the spectrophotometric method described by Placer et al., (1966), reduced glutathione (GSH) level was determined according to the method described by Sedlak and Lindsay (1968), glutathione peroxidase (GSH-Px) activity was determined as described by Lawrence et al., (1976), and catalase (CAT) activity was determined according to the spectrophotometric method described by Aebi (1984).

For histological analyses, 1-cm samples were taken from the duodenal region of the digestive tract from all slaughtered animals and fixed in 10% buffered neutral formalin solution for 24 hours (Xu et al., 2003). The tissues were dehydrated by passing through 70%, 80%, 96% and 100% alcohol series for routine tissue processing procedures. Subsequently, the tissues were treated with xylol for transparency and fixed in soft paraffin. Then, paraffin blocks were prepared from the tissues embedded in hard paraffin, and 5-µm thick sections were taken with a microtome (Thermo Scientific, HM325) for morphometric analysis. Haematoxylin-Eosin (H-E) staining technique was applied to perform morphometric measurements (villus height, villus width and crypt depth) of duodenal tissues. Preparations covered with Entellan were examined at 20x magnification with a Zeiss Axiolab 5 microscope (Carl Zeiss Ltd.) equipped with an Axiocam 208 colour digital camera with Zeiss ZEN 3.5 blue edition software, and their measurements were taken and statistical analyses were performed. Figure 1 shows the measured parameters of the duodenum section stained with Haematoxylin-Eosin (H-E) under light microscope. All these procedures were performed in the laboratories of the Department of Histology and Embryology, Faculty of Veterinary Medicine, Fırat University.

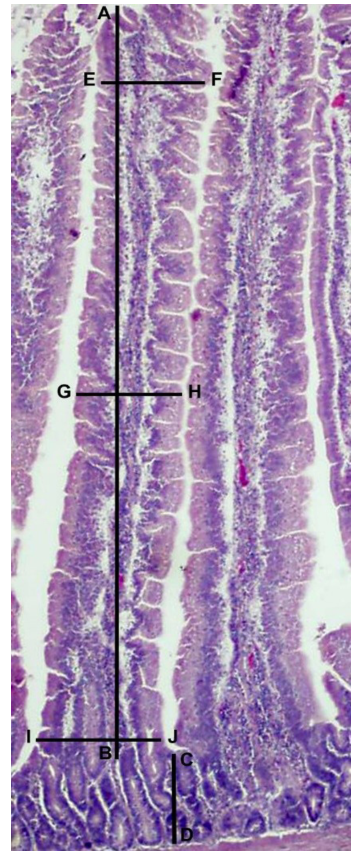

Figure 1. Hematoxyline-eosin (H&E) stained duodenum section shows the measured parameters **[AB]:** Villus height, **[CD]:** Crypt depth, **[EF]:** Villus upper width, **[GH]:** Villus middle width and **[IJ]:** Villus bottom width (Magnification: x200)

Statistical Analysis

After normality analysis (Shapiro-Wilk) was performed on the data obtained at the end of the study, analysis of variance was used to compare the groups for blood analysis, oxidative stress and antioxidant parameters of liver tissue and histological analysis was run for duodenal tissue. Duncan's test was used for further analysis. SPSS packaged software was used for the analysis (Kalaycı, 2006). Data were presented as Mean ± Standard error of the mean and differences were considered as significant at the level of P≤0.05.

RESULTS

Table 4 shows the blood parameters (glucose, triglyceride, total cholesterol, VLDL cholesterol, ALT, AST, uric acid, and creatine) of the broiler chickens. When Table 4 is analysed, it was found that the lowest glucose level was 185.13 mg/dL in the H-1

Table 4. The effect of jujube fruit added to mixed feed on blood parameters in broiler chickens, ($\bar{x} \pm S_{\bar{x}}$) (n=8)

	Jujube, %				
Item	**Control**	**0.5**	**1**	**2**	**P**
Glucose (mg/dL)	227.00±9.05^{a}	207.50±7.89^{ab}	185.13±6.33^{b}	188.25±6.17^{b}	*
Triglycerides (mg/dL)	83.25±11.40^{a}	61.13±5.37^{b}	51.13±6.39^{b}	54.88±5.78^{b}	*
Total cholesterol (mg/dL)	128.13±8.17^{a}	118.13±6.22^{ab}	113.13±10.81^{b}	112.88±7.19^{b}	*
VLDL cholesterol (mg/dL)	16.65±2.28^{a}	12.23±1.07^{b}	10.23±1.28^{b}	10.98±1.16^{b}	*
ALT, U/L	5.75±0.45^{a}	5.13±0.40^{b}	5.13±0.35^{b}	5.38±1.02^{ab}	*
AST, IU/L	603.50±29.19^{a}	513.88±66.56^{b}	455.63±37.47^{c}	535.25±62.75^{ab}	*
Uric acid, mg/dL	8.63±0.60^{a}	7.75±0.70^{a}	7.38±0.55^{ab}	5.88±0.30^{b}	*
Creatine, mg/dL	0.10±0.00^{b}	0.11±0.01^{ab}	0.11±0.01^{ab}	0.12±0.01^{a}	*

* : P<0.05, $^{a-c}$: The difference between the values expressed with different letters in the same row is significant.

VLDL: Very Low Density Lipoprotein, **ALT:** Alanine Aminotransferase, **AST:** Aspartate Aminotransferase group, followed by 188.25 mg/dL in the H-2 group, 207.50 mg/dL in the H-0.5 group and 227.00 mg/dL in the control group ($P<0.05$). The highest triglyceride, total and VLDL cholesterol levels were detected in the control group ($P<0.05$). While the H-2 group had the lowest uric acid level, the control group had the lowest creatine level ($P<0.05$). The highest ALT and AST levels were determined in the control group ($P<0.05$). Table 5 shows antioxidant (malondialdehyde, reduced glutathione, glutathione peroxidase and catalase) parameters of liver tissue of broiler chickens. Accordingly, the lowest malondialdehyde (MDA) level ($P<0.001$) was found in the H-2 group and the lowest CAT level ($P<0.05$) was detected in the control group. GSH levels were higher in H-1 and H-2 groups than in H-0.5 and control groups ($P<0.01$). There was no statistical difference between the groups in terms of GSH-Px ($P>0.05$). Table 6 shows the morphological structure of the duodenum of broiler chickens and Figure 1 shows the measured parameters of the duodenal section stained with haematoxylin-eosin. When Table 6 was analysed, it was found that the highest values in terms of villus height, crypt depth, villus bottom width and villus edge width were observed in the H-1 and H2 groups ($P<0.001$). In the parameter of villus height/crypt depth, contrary to the above, the lowest levels were determined in the H-1 and H2 groups ($P<0.001$). In the villi upper width parameter, the highest value was observed in the H-2 group ($P<0.001$).

DISCUSSION

In the poultry sector, the use of feed additives is an important strategy to increase animal productivity, prevent diseases and ensure better feed utilization (Pirgozliev et al., 2019).

When the blood parameters of broiler chickens were examined in the present study, it was found that glucose, triglyceride, total and VLDL cholesterol levels lowered as a result of addition of jujube

Table 5. The effect of jujube fruit added to mixed feed on oxidative stress and antioxidant parameters in liver tissue of broiler chickens, ($\bar{x} \pm S\bar{x}$) (n=8)

	Jujube, %				
Item	**Control**	**0.5**	**1**	**2**	**P**
MDA, nmol/g	14.72±0.36^{a}	13.81±0.47^{a}	12.48±0.46^{b}	10.81±0.18^{c}	***
GSH, nmol/g	2.08±0.07^{b}	2.06±0.05^{b}	2.47±0.10^{a}	2.32±0.09^{a}	**
GSH-Px, IU/g protein	38.22±0.46	37.40±0.97	38.31±0.53	37.62±0.87	NS
CAT, KU/L	43.09±0.84^{b}	45.18±0.80^{ab}	46.94±0.70^{a}	45.75±0.81^{a}	*

NS: P>0.05, * : P<0.05, ** : P<0.01, *** : P<0.001, $^{a-c}$: The difference between the values expressed with different letters in the same row is significant.

MDA: Malondialdehyde, **GSH:** Reduced Glutathione, **GSH-Px:** Glutathione Peroxidase, **CAT:** Catalase

Table 6. The effect of jujube fruit added to mixed feed on duodenal morphology in broiler chickens, ($\bar{x} \pm S\bar{x}$) (n=8)

	Jujube, %				
Item	**Control**	**0.5**	**1**	**2**	**P**
Villus Height	156.58±4.09^{c}	174.08±2.94^{b}	208.30±1.31^{a}	206.18±1.29^{a}	***
Crypt Depth	21.60±1.20^{c}	26.98±0.96^{b}	39.18±0.80^{a}	40.88±0.57^{a}	***
Villus Upper Width	17.25±0.94^{d}	24.00±0.87^{c}	30.28±0.51^{b}	33.53±0.62^{a}	***
Villus Bottom Width	21.78±0.88^{c}	30.88±1.15^{b}	38.85±0.64^{a}	40.80±0.54^{a}	***
Villus Edge Width	20.98±0.95^{c}	29.70±0.97^{b}	40.20±0.54^{a}	40.43±0.52^{a}	***
Villus Height/ Crypt Depth	8.39±0.62^{a}	6.86±0.32^{b}	5.42±0.13^{c}	5.08±0.07^{c}	***

*** : P<0.001, a-d: The difference between the values expressed with different letters in the same row is significant.

to the mixed feed, the lowest uric acid level was determined in the H-2 group and the lowest creatine level and the highest ALT and AST levels were determined in the control group. Furthermore, jujube fruit added to mixed feed lowered glucose levels. Although the mechanism of jujube fruit in lowering the glucose level is not known exactly, a study reported that jujube extracts affected pancreatic beta cells, resulting in elevating the insulin level and as a result, the hypoglycaemic effect takes place by increasing the insulin level (Gülay, 2013). Basiriyan et al., (2022) conducted a study to investigate the effects of jujube and bird spindle powder addition to broiler compound feed on growth performance, blood metabolites and meat quality under high temperature conditions and reported that jujube and bird spindle powder added to the compound feed significantly reduced blood glucose. All these results indicate that jujube fruit has a hypoglycaemic effect and are consistent with the findings obtained in this study on glucose. Cholesterol is essential for life and is distributed throughout the body (brain, nerves, heart, intestines, muscles and liver). While low levels of cholesterol in the blood are sufficient, excessive levels can cause problems for heart and body health (Dirican et al., 2017). In this study, it was observed that total and VLDL cholesterol levels lowered as a result of jujube addition to mixed feed. In a study, it was found that cholesterol levels were lower in all groups to which Ziziphus mauritiana leaf extract (AZL) was added compared to the control group and this was attributed to the vitamin and ascorbic acid (vitamin C) contained in the components of AZL (Abdulameer et al., 2017). Indeed, in another study, it was reported that cholesterol in blood decreased with vitamin C (Sarandol et al., 2023). A study investigating the effects of jujube fruit supplementation at different rates (0, 2.5, 5 and 7.5%) into mixed feeds on growth performance parameters, carcass yield and some biochemical parameters of broiler chickens reported that the abdominal fat of jujube-fed animals was low and serum total cholesterol level lowered with increasing jujube level (Gholezoo et al., 2015). Flavonoids are reported to reduce plasma levels of low-density lipoprotein (LDL) through an upregulation of the LDL receptor (Bjune et al., 2024). As a matter of fact, total and VLDL cholesterol levels in the groups with jujube may have been found to be lower than the control group by this mechanism due to the flavonoides in the structure of jujube fruit. Triglycerides represent a heterogeneous group of lipids formed by the esterification of three fatty acids with glycerol. Since they are insoluble in water, they are transported in lipoproteins in the blood. In this study, the highest triglyceride level was found in the control group and jujube added to the mixed feed lowered the triglyceride level. Indeed, a previous study reported that jujube extract could potentially prevent obesity in rats and also inhibit lipid accumulation and glycerol-3-phosphate dehydrogenase activity without impairing cell viability (Kubota et al., 2009). Glycerol-3-phosphate dehydrogenase enzyme is necessary for triglyceride production. As a matter of fact, it was determined that the addition of alfalfa flavonoide into broiler mixed feeds lowered the serum triglyceride level depending on the dose increase (Ouyang et al., 2016). In contrast to this study, it was reported that 45 and 90 mg/kg levels of jujube leaf extract added to compound feed increased serum triglyceride concentration in laying hens (Kılınç et al., 2020). Similarly, El-Maaty et al., (2018) found that jujube leaf extract in broiler diets increased triglyceride concentration compared to the control group. On the other hand, Gholezoo et al., (2015) investigated the effects of jujube fruit supplementation at different rates (0, 2.5, 5 and 7.5%) in mixed diets on growth performance parameters, carcass yield and some biochemical parameters of broiler chickens and reported that jujube supplementation had no effect on serum triglyceride level. Alanine aminotransferase (ALT) and aspartate aminotransferase (AST) are the two most important enzymes distributed in the liver cytoplasm and mitochondria. Elevated levels of these two enzymes are generally accepted as an indicator of liver cell damage (Anurag et al., 2015). Under normal conditions, the activities of these two enzymes in serum are low. However, it is suggested that these enzymes gradually enter the blood after hepatocellular damage (Ezhilarasan, 2018). Therefore, determination of the activities of these two enzymes in serum is sufficient for the prognosis of acute liver injury. In this study, jujube fruit added to compound feed lowered ALT and AST levels compared to the control group. In addition to the study in which the addition of jujube decreased ALT and AST (Liu et al., 2022), there is a study in which ALT level was found to be higher in the jujube group (El-Maaty et al., 2018) and a study reporting that jujube addition did not affect AST level in broiler chickens (Kılınç et al., 2020). Generally, laboratory-based criteria of liver and kidney toxicity are explained by ALT and/or alkaline phosphatase (ALP) and blood urea nitrogen (BUN) and/or creatinine values, respectively. In the present study, the lower values of both ALT and AST levels compared to the control group can be interpreted as an indication that the doses we used did not have a negative effect on the liver in animals. Uric acid (UA) is the end product of exogenous and endogenous purine metabolism and its overproduction or under-excretion can lead to hyperuricemia (Yamauchi and Ueda, 2008). Normally, serum uric acid (SUA) levels are balanced between hepatic production and renal and intestinal excretion (El Ridi and Tallima, 2017). In this study, the highest serum uric acid level was detected in the control group and the serum uric acid level tended to decrease depending on the level of jujube added into the mixed diet. Natural compounds derived from plants have a significant potential to lower blood uric acid. Flavonoids, especially flavones and flavonols, as well as saponins, terpenoids and alkaloids, show uric acid-lowering effects. These compounds can reduce uric acid in the blood by inhibiting xanthine oxidoreductase and regulating uric acid transporters (Feng et al., 2022). The decrease in uric acid level in this study can be attributed to the flavonoids in its structure. As a matter of fact, uric acid data in studies conducted with herbal products show similarities with the findings of this study (Çiftçi et al., 2016, El-Shenway and Ali, 2016). Creatinine is the waste product of the breakdown of a phosphate compound called creatine produced in muscle and protein metabolism, which the body uses to provide energy to the muscles. The kidneys keep the creatinine level in the blood within the normal range. In this study, the highest creatinine level was found in the H-2 group. Different results were obtained on serum creatinine level in studies conducted with thyme oil addition into broiler mixed diets. As a matter of fact, it was observed that there was a linear decrease in serum creatinine level of quail as a result of 150, 300 and 450 mg/kg thyme oil addition to compound feed compared to the control group (Gumus et al., 2017). On the other hand, it was determined that there was no difference in serum creatinine concentration in broiler chickens consuming 1 g/kg thyme powder (Ragaa et al., 2016), but serum creatinine concentration increased significantly in broiler chickens fed with 10 g/kg thyme powder compared to untreated broiler chickens (Tayeb et al., 2019). The creatinine level obtained in this study may have increased depending on the dose used.

Stress in poultry is one of the important factors that increase the production of mitochondrial reactive oxygen species (ROS) in tissues (Kikusato et al., 2016). ROS accumulate in cells and cause irreversible damage to molecules such as lipids, proteins and DNA, leading to cellular disorders (Kikusato et al., 2016, Azad et al., 2013). MDA is the main end product of lipid peroxidation and is often used to determine oxidative damage. In parallel with the increase in the level of jujube used in the study, MDA level lowered, while GSH and CAT levels elevated. There was no effect of jujube on GSH-Px level. This may be attributed to the fact that jujube fruit has strong antioxidant properties due to its abundant polyphenol content such as protocatechuic, gallic, and chlorogenic acids. Yang et al., (2023) investigated the effects of dried jujube fruit powder (DJFP) supplementation into broiler mixed diets on growth performance, antioxidant stability, meat composition and quality and found that DJFP supplementation significantly increased total antioxidant capacity (T-AOC), SOD, CAT and GSH-Px activities in breast muscle. Similarly, it was reported that the addition of 1% jujube into the mixed feed improved the antioxidant enzyme activities in the muscles of Japanese quails (Cellat et al., 2022). Another study investigated the effects of phytogenic additives (jujube, castor nut, blackberry and pomegranate leaf extracts) added into the diet as an alternative to antibiotics on performance and some blood parameters in broiler chickens. The results of the study revealed that although there was no significant difference between the groups in terms of plasma GSH, SOD level was higher in the groups fed with phytogenic additives than in the control group (El-Maaty et al., 2018).

The small intestine is the main site for digestion and absorption of nutrients. Long villi allow for a healthy digestive system and high absorption of nutrients in poultry (Alfaro et al., 2007, Sims et al., 2004). Villi and crypts in the small intestine are functional units that play important roles in digestion and absorption. Increasing the height of the villi increases the activation of digestive enzymes by expanding the total luminal villus absorption area, thus promoting more efficient absorption of nutrients on the villus surface (Cera et al., 1988). In crypts, epithelial cell proliferation occurs. Therefore, crypt development directly affects villus development and intestinal absorption surface and thus efficiency (Geyra et al., 2001). In the present study, duodenal villus height, villus upper width, villus lower width, villus edge width and crypt depth increased depending on the level of jujube added into the mixed diet. In their study conducted to determine the effects of jujube fruit added into the feed on performance, small intestinal histo-morphometry, oxidative stress and carcass parameters in Japanese quails raised at two settlement densities (14, 21 quails/cage), Cellat et al., (2022) found that the addition of jujube fruit increased villus height and crypt depth. Among some studies conducted on broiler chickens; villus length, crypt depth and goblet cell number increased in the study by Köksal et al., (2012) using an essential oil mixture (Origanum vulgare, Thymus vulgare, garlic, anise, and fennel) at a dose of 0.75 g/kg, in the study by Yıldız (2007) using an essential oil mixture (carvacrol, thymol, rosmarinic) at a dose of 1 g/kg, Jerzsele et al., (2012) with 1.5 g/kg essential oil mixture (ginger and carvacrol), in the study by Demir et al., (2005) using origanum powder and thyme powder at a dose of 1 g/kg, in the study by Garcia et al., (2007) using plant extract (origanum, rosemary and sage) at a dose of 5000 ppm, in the study by Hong et al., (2012) using essential oil mixture (thyme, anise and citrus peel) at a dose of 125 ppm, and in the study by Silva et al., (2009) using 0.5 and 1 g/kg origanum oil. Therefore, the findings of the present study support the effects of herbal products on broiler chickens.

CONCLUSION

Consequently, it was concluded that jujube fruit used in this study can be easily used in poultry feeding due to its positive effects on blood parameters, liver tissue antioxidant parameters and duodenal mucosa in broiler chickens, especially at the level of 1%.

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