

Effects of quebracho condensed tannin on growth performance, digestibility, health parameters and mortality in chukar partridges chicks

Hasan Tarık EŞKİ¹ (✉), Nurettin GÜLŞEN²

¹ Bahri Dagdas International Agricultural Research Institute, Konya, Türkiye

² Selçuk University Veterinary Faculty, Animal Nutrition and Nutritional Disorders, Selcuk University, Konya, Türkiye

✉ Corresponding author: hsntarik@gmail.com

Received: September 26, 2024; accepted: December 19, 2024

ABSTRACT

This study aimed to determine the effects of quebracho (*Schinopsis balansae*) condensed tannin addition to diet on growth performance, feed intake, digestibility, relative carcass and internal organ weights, mortality and the number of *Eimeria* oocysts in feces of chukar partridge (*Alectoris chukar*) chicks. Partridge chicks were allocated into 5 treatments and 6 replications, containing 0% (control), 0,25%, 0,5%, 1% and 2% tannin. Twelve chicks were placed in each replication and 72 chicks in each group, having a total of 360 partridge chicks. The experiment lasted 12 weeks, with the first 6 weeks of the starter period and the last 6 weeks of the grower period. It was observed that the effects of quebracho condensed tannin addition to diet did not affect performance, relative carcass, liver, gizzard, stomach and small intestine weights were found to be insignificant, but it reduced the large intestine weight ($P < 0,05$). Differences between groups in nutrient and mineral digestibility were found to be insignificant but calcium digestibility was found to be decreased in animals given 2% tannin ($P < 0,01$). Tannin addition to diet does not affect the cecum and feces parameters. It has been observed that tannin reduces the oocyte counts in the 1% and 2% treatments. Condensed tannin did not affect the amount of plasma immune proteins, but viability was observed to increase significantly in the 2% tannin treatment ($P < 0,05$). As a result, tannin addition to the diet increased the viability of partridge chicks without suppressing growth performance and digestibility, and it was concluded that 2% quebracho condensed tannin could be used in partridge chick rations to decrease mortality.

Keywords: partridge, condensed tannin, digestion, immunity, FCR

INTRODUCTION

Poultry meat and eggs play a significant role in meeting the animal protein needs of humans (Scholten et al., 2013). The continuous increase in global population, improvement in consumption levels, and accelerated urbanization have consistently boosted market demand for poultry products. In recent years, with the growing interest in game bird meat, the production of quail, partridge, and pheasant has also gained popularity (Kokoszynski et al., 2013). The chukar partridge is one of the most popular game birds, inhabiting a wide geographic range from the Balkans to Far East Asia (Panayides et al., 2011). However, due to overhunting, habitat destruction,

and environmental pollution, the partridge population has been reported to be declining, making intensive farming conditions increasingly important (Karadas et. al., 2017).

From the 1950s to the 2000s, growth-promoting antibiotics were added to animal feed to enhance growth performance in farm animals. The addition of antibiotics to feed to improve microbiota and the immune system in animals has been banned due to the global health risks associated with antibiotic resistance, leading to the search for alternative approaches (Redondo et al., 2014). Among the substances that could be used as alternatives to antibiotics, phytochemicals derived from plants have gained prominence in research. Among these, tannins

are particularly notable in the poultry sector, where they are most commonly used to replace growth-promoting antibiotics due to their antimicrobial and growth-enhancing properties in broilers. Tannins are secondary metabolites found in plants in large quantities, following cellulose, hemicellulose, and lignin (Chowdhury et al., 2004). Tannins are present in various parts of plants and trees, such as leaves, stems, fruits, and bark, and they exhibit different physical and chemical properties. Tannins are generally classified into three groups: hydrolyzable tannins (HT), condensed tannins (CT), and phlorotannins. While hydrolyzable and condensed tannins are plant-derived, phlorotannins are obtained from brown algae (Chung et al., 1998).

Condensed tannins, also known as proanthocyanidins, are non-hydrolyzable oligomeric and polymeric compounds with molecular weights ranging from a few hundred to several thousand Daltons. Their multiple phenolic hydroxyl groups form complexes with proteins, metal ions, and other macromolecules like polysaccharides (Schofield et al., 2001). Unlike hydrolyzable tannins, condensed tannins are resistant to hydrolysis, making their bioavailability in the digestive tract low. This is due to their high molecular weight, which leads to very low absorption in the intestines. Condensed tannins are the most abundant tannins found in fruits (such as blackberries, apples, pears), legume forages (such as lentils, cowpeas, chickpeas, Mexican beans), nuts, green and red grapes (in grape juice and wines), trees, and stems. They are particularly abundant in plants like bird's-foot trefoil (*Lotus corniculatus*), found in legume-rich pastures (Degen et al., 1995).

In the past, tannins were considered harmful to monogastric animals due to their negative effects on nutrient digestibility, nitrogen retention, and productivity traits (Garcia et al., 2004). However, recent studies have shown that the effects of tannins vary depending on the animal species, the amount used, the duration of use, and the chemical structure of the tannins (Redondo et al., 2022). Tannins added to the diet have been shown

to reduce gastrointestinal parasites in poultry (Marzoni et al., 2020) and improve gut bacterial colonization through their antimicrobial effects (Scalbert, 1991). Although there have been studies on the effects of quebracho condensed tannins in ducks (Castillo et al., 2020), pheasants (Marzoni, et al., 2005b), and chickens (Marzoni et al., 2020), information on their effects in chukar partridges is limited. This study aims to investigate the effects of different levels of quebracho condensed tannins on growth performance, digestibility, and certain health parameters in chukar partridge chicks.

MATERIAL AND METHODS

Ethical Statement

This research project was conducted with the ethical approval of the Selçuk University Faculty of Veterinary Medicine Animal Production and Research Center Ethics Committee (SÜVDAMEK) as per decision number 2021/120, dated 17.11.2021, during the meeting numbered 2021/10.

Dietary treatments, birds, and management

A total of 360 mixed-gender chukar partridge chicks were divided into five different groups based on tannin levels in feed, with each group consisting of six replicates (12 chicks/replicate). The subgroups were randomly allocated to cages of 0.5 m² each. Throughout the experiment, the partridge chicks had ad-libitum access to feed and water. The experimental room was heated with thermostatic radiators, starting at a temperature of 35 °C, which was gradually reduced by 1 °C per week until it reached 24 °C. The experiment was conducted in two periods: the starter phase and the grower phase. The partridge chicks were fed a starter diet during the first 6-week period, followed by a grower diet in the subsequent 6-week period. The compositions of the diets for the starter and grower phases are shown in Table 1. The nutritional requirements of the diets were determined according to (Gülşen et al., 2010) and were prepared to be isocaloric and isonitrogenous.

Table 1. Composition and nutrient levels of starter (0-5 weeks) and grower diets (6-12 weeks)

Feed ingredient (%)	Starter period					Growth period				
	T0	T1	T2	T3	T4	T0	T1	T2	T3	T4
Corn	24.3	24	23.3	21.7	19.3	32.5	32	31.5	30.5	28.7
Wheat	19	19.3	19.8	21	22	23.2	23.7	24	24.6	25.8
SBM (48% CP)	41	41.2	41.6	42.1	43.2	30	30.1	30.5	31.2	32.9
CSM (32% CP)	4.5	4	3.5	2.5	1.25	4.5	4	3.5	2.5	0
Vegetable oil	2.75	2.8	2.9	3.1	3.7	1.4	1.5	1.6	1.8	2.15
Razmol	5	5	5	5	5	5	5	5	5	5
Tannin	-	0.25	0.5	1	2	-	0.25	0.5	1	2
DCP	1.4	1.4	1.4	1.4	1.4	1.6	1.6	1.6	1.6	1.6
Limestone	1.4	1.4	1.4	1.4	1.4	1.35	1.35	1.35	1.35	1.35
Salt	0.33	0.33	0.33	0.33	0.33	0.35	0.35	0.35	0.35	0.35
Vit.-Min. premix*	0.1	0.1	0.1	0.1	0.1	0.1	0.1	0.1	0.1	0.1
L-Lysine	0.22	0.22	0.22	0.22	0.22	-	-	-	-	-
Calculated nutrient levels										
Dry Matter (%)	89.7	89.4	89.2	88.8	87.9	89.50	89.50	89.50	89.40	89.00
Metabolizable energy (kcal/kg)	2804	2802	2800	2801	2802	2768	2766	2767	2768	2767
Crude protein (%)	26	26	26	26	26	23.00	23.00	23.00	23.00	23.00
Calcium (%)	1.01	1	1	1	1	0.90	0.90	0.90	0.90	0.90
Phosphorus (%)	0.46	0.46	0.46	0.46	0.46	0.43	0.43	0.43	0.43	0.43
Sodium (%)	0.17	0.17	0.17	0.17	0.17	0.17	0.17	0.17	0.17	0.17
Chlorine (%)	0.24	0.24	0.24	0.24	0.24	0.24	0.24	0.24	0.24	0.24
Methionine + Cysteine (%)	0.85	0.85	0.85	0.85	0.85	0.79	0.79	0.79	0.79	0.79
Lysine (%)	1.61	1.61	1.61	1.62	1.63	1.35	1.35	1.35	1.36	1.37
Threonine (%)	0.97	0.97	0.97	0.98	0.98	0.86	0.86	0.86	0.86	0.86
Tryptophan (%)	0.38	0.38	0.38	0.38	0.39	0.36	0.36	0.36	0.37	0.37
Analyzed nutrient levels										
Dry matter (%)	91.6	91.5	92	91.6	91.6	91.7	91.4	91.5	91.3	91.6
Crude ash (%)	5.17	5.06	4.7	4.94	5.85	4.63	4.59	5.07	5	4.65
Crude fat (%)	4.04	3.47	4.04	3.74	4.07	4.07	4.13	4.22	4.24	4.47
Crude Protein (%)	25.7	25.6	25.4	25.9	25.8	22.7	22.6	22.8	22.7	22.9
ME energy (kcal/kg)	2,904	2,890	2,922	2,916	2,918	2,901	2,893	2,913	2,912	2,904

* Vitamin-mineral premix (per kg feed): 12,500 IU vit A, 2,500 IU vit D3, 30 mg vit E, 10 mg niacin, 1,600 mg Ca, 1,300 mg P, 0.4 mg Mg, 3.2 mg Zn, 3.0 mg Mn, 8 mg Cu, 0.4 mg I, 0.1 mg Co, 0.1 mg Se, 500 mg NaCl, 300 mg NaHCO₃

Both the starter and grower diets were supplemented with condensed tannin at levels of 0% (T0), 0.25% (T1), 0.5% (T2), 1% (T3), and 2% (T4). A commercial product containing 100% condensed tannin in powder form (MGM-S, Buenas Aires, Argentina) was used as the tannin source.

Growth performance

The partridges were weighed every two weeks to calculate the body weight and daily weight gain (DWG) per bird. Feed intake was determined weekly by weighing the remaining feed, and feed conversion ratios (FCR) were calculated for each group by dividing the total feed consumed during each period by the weight gain for that period. Mortality rates were monitored daily.

Determination of carcass and organ weights

On the final day of the experiment, the partridges were weighed before slaughter, and the weights of the liver, proventriculus, gizzard, and filled intestines were recorded. These digestive system parts were emptied, rinsed with cold (+4 °C) saline solution, dried with filter paper, and weighed again to determine their empty weights. Relative organ weights were calculated using the following formula (Mossa et al., 2015):

$$\text{Relative organ weight} = (\text{Organ weight} / \text{Body weight}) \times 100$$

The cecum content was collected for pH, volatile fatty acids (VFA), lactic acid, and ammonia level determination. After the feathers were mechanically removed and the internal organs extracted, the head, neck, and feet were cut off, and the carcass weight was recorded.

Nutrient analyses and digestibility determination

The dry matter, ash, crude protein, and crude fat contents of the diets were analyzed according to AOAC (2003), and the crude fiber content was analyzed according to Van Soest et al. (1991) by Ankom 200 device. Manure was collected from trays under the cages for three consecutive days and dried in an oven at 65 °C for 48 hours, and the dry matter content of the manure was determined. A composite sample of the manure

was then taken for nutrient analysis. The digestibility of the nutrients in the diets was calculated using the acid-insoluble ash (AIA) method, with digestibility coefficients determined using the following formula (Diana et al., 2022):

$$\text{Digestibility of nutrients in the diets (\%)} = 100 - [100 \times (\text{AIA}_{\text{diet}} \times \text{Nutrient}_{\text{excreta}}) / (\text{AIA}_{\text{excreta}} \times \text{Nutrient}_{\text{diet}})]$$

The amounts of calcium, phosphorus, and magnesium in the feed and feces were determined using Inductively Coupled Plasma-Atomic Emission Spectrometry (Soltanpour et al., 1996) (ICP-AES, Varian-Vista, Australia). Feces scoring was done according to the method reported by Swanson et al. (2002).

Protozoa load

At the end of the experiment, fresh fecal samples were randomly taken from the collection trays and examined for *Eimeria* spp. oocysts using the McMaster egg counting method (Chandrawathani et al., 2015):

$$\text{Oocyst count per gram of feces} = (\text{Egg count in chamber 1} + \text{Egg count in chamber 2}) \times 50$$

Determination of volatile fatty acids (VFA), pH, ammonia, and lactic acid in cecal contents

The pH values of the samples were measured using a digital pH meter (HI 8314, Hanna Instruments, Portugal). Lactic acid levels were determined using a spectrophotometric method reported by Borshchevskaya et al. (2016). To determine the concentrations of acetic acid (AA), propionic acid (PA), butyric acid (BA), and total VFA measurements were made using a gas chromatograph (Agilent 6890N GC, USA) equipped with a DB-FFAP capillary column (30 m × 320 µm × 1.00 µm; Agilent J&W GC column) and an FID detector (Sigma-Aldrich 1998). The "Supelco, Volatile Fatty Acid Mix CRM46975" mixture was used as a standard.

Blood analyses

At the end of the experiment, blood samples were collected from 30 randomly selected partridges (one from each replicate) into tubes containing a gel activator for IgG,

IgE, and IgA analysis. The blood samples were centrifuged at 1500 rpm for 10 minutes to obtain serum. IgG, IgE, and IgA analyses were performed using the Abbott Architect Ci 8200 device with a turbidimetric method (Tietz, 1995).

Statistical analysis

Statistical analyses were performed using SPSS (V25.0). Normality tests were conducted using the Shapiro-Wilk test, and homogeneity was assessed using Levene's test. For variance analysis, one-way ANOVA was used, and for non-normally distributed data, the Kruskal-Wallis test was employed. Differences between groups were determined using the Tukey test. Survival rates were analyzed using the chi-square test. Differences between groups were considered significant at $P < 0.05$.

RESULTS

Performance

No statistically significant differences were found between the groups in terms of live weight gain and cumulative live weight gain, cumulative feed intake, and feed conversion ratio during the initial period (0-5 weeks) and the growing period (6-12 weeks; Table 2).

Effects of tannin addition to diet on digestibility

Digestibility data for dry matter, crude protein, crude fat, calcium, phosphorus, and magnesium according to increasing tannin levels are shown in Table 3. According

to the table, the differences between the groups were insignificant ($P > 0.05$). However, the differences between the groups were significant regarding calcium digestibility ($P < 0.05$).

Carcass and internal organ weights

According to the results given in Table 4, the differences between the groups were statistically insignificant in terms of carcass, liver, gizzard full and empty weights, stomach full and empty weights, small intestine empty weight, and large intestine full weight. However, the differences between the groups were statistically significant in terms of large intestine empty weight ($P < 0.05$).

Cecum and Fecal Parameters

No significant differences were found between the groups in terms of cecal lactic acid concentration, pH, NH_3 , SCFA, fecal dry matter, fecal score, and fecal NH_3 concentration ($P > 0.05$) (Table 5).

Fecal Eimeria oocyst count, plasma immunoglobulin levels, and survival rate

The oocyst counts in the feces of *Eimeria* spp. showed significant differences among the groups ($H(4) = 15.082$, $P < 0.05$). The serum IgA, IgG, and IgE levels are presented in Table 6. It was determined that the differences between the groups were insignificant ($P > 0.05$), with the values

Table 2. Cumulative live weight gains of partridges during the initial and growing periods according to the amount of tannin added to the diet (g)

	T0 ($\bar{X} \pm S_x$)	T1 ($\bar{X} \pm S_x$)	T2 ($\bar{X} \pm S_x$)	T3 ($\bar{X} \pm S_x$)	T4 ($\bar{X} \pm S_x$)	P Value
0-5 weeks	176 ± 4.08	189 ± 4.68	179 ± 7.63	179 ± 10.11	178 ± 5.8	0.69
6-12 weeks	192 ± 6.4	166 ± 11.18	202 ± 11.6	188 ± 13.77	203 ± 6.85	0.12
Cumulative LWG	368 ± 4.47	355 ± 15.28	381 ± 9.1	367 ± 12.52	381 ± 6.32	0.2
Avg. daily gain	4.30 ± 0.14	4.17 ± 0.14	4.41 ± 0.14	4.16 ± 0.14	4.45 ± 0.14	0.38
Cumulative FI	2417 ± 102	2328 ± 122	2349 ± 67.7	2340 ± 61.6	2382 ± 44.4	0.95
FCR (g feed/g LWG)	6.56 ± 0.23	6.57 ± 0.28	6.18 ± 0.18	6.41 ± 0.27	6.27 ± 0.17	0.69

*T0: Control, T1: 0.25% tannin, T2: 0.5% tannin, T3: 1% tannin, T4: 2% tannin

found to be numerically close to each other. The study observed that the survival rate of the group containing 2% tannin was significantly higher than that of the control group and the groups containing 0.5% and 1% tannin ($P < 0.05$). However, it was found that there was only a numerical difference between the group containing 2% tannin and the group containing 0.25% tannin. The mortality rates of the other groups were found to be numerically quite close to each other (Table 6).

Table 3. Effects of increasing tannin levels in the diet on the digestibility of dry matter, organic matter, crude protein, crude fat, calcium, phosphorus, and magnesium (%)

Digestibility %	T0 ($\bar{X} \pm S_x$)	T1 ($\bar{X} \pm S_x$)	T2 ($\bar{X} \pm S_x$)	T3 ($\bar{X} \pm S_x$)	T4 ($\bar{X} \pm S_x$)	P Value
Dry matter (DM)	78.6 \pm 1.14	77.4 \pm 0.84	76.5 \pm 1.47	76.6 \pm 0.1	76.3 \pm 1.35	0.58
Organic matter (OM)	80.6 \pm 1.04	80.6 \pm 0.69	79.6 \pm 1.29	79.7 \pm 0.16	77.6 \pm 0.45	0.14
Crude protein (CP)	58.1 \pm 2.4	55.3 \pm 2.02	49.1 \pm 3.34	52.0 \pm 1.02	51.1 \pm 2.69	0.18
Crude fat (CF)	92.4 \pm 0.66	91.1 \pm 0.93	90.0 \pm 0.51	92.2 \pm 0.51	90.4 \pm 2.25	0.51
Calcium (Ca)**	55.5 \pm 3.29 ^a	47.0 \pm 2.53 ^{ab}	49.2 \pm 3.08 ^{ab}	47.3 \pm 3.04 ^{ab}	37.2 \pm 2.58 ^b	0.01
Phosphorus (P)	50.8 \pm 3.41	50.4 \pm 1.34	50.2 \pm 3.16	49.3 \pm 2.02	44.4 \pm 2.06	0.39
Magnesium (Mg)	42.1 \pm 5.13	39.8 \pm 2.54	38.0 \pm 5.06	38.0 \pm 1.06	32.4 \pm 3.42	0.49

** Different letters indicate statistically significant differences between groups ($P < 0.05$)

Table 4. Comparison of carcass and relative organ weights according to the level of tannin added to the diet (g)

Visceral organs	T0 ($\bar{X} \pm S_x$)	T1 ($\bar{X} \pm S_x$)	T2 ($\bar{X} \pm S_x$)	T3 ($\bar{X} \pm S_x$)	T4 ($\bar{X} \pm S_x$)	P Value
Carcass	228 \pm 5.99	241 \pm 12.07	225 \pm 30.32	235 \pm 16.8	257 \pm 9.31	0.7
Liver	2.42 \pm 0.15	2.26 \pm 0.17	2.23 \pm 0.08	2.7 \pm 0.34	2.15 \pm 0.13	0.31
Gizzard	2.8 \pm 0.16	2.76 \pm 0.12	2.58 \pm 0.1	2.92 \pm 0.15	2.68 \pm 0.12	0.46
Stomach	0.39 \pm 0.05	0.3 \pm 0.02	0.33 \pm 0.02	0.35 \pm 0.02	0.36 \pm 0.01	0.24
Small intestine	1.42 \pm 0.14	1.46 \pm 0.07	1.5 \pm 0.05	1.76 \pm 0.06	1.42 \pm 0.07	0.07
Large intestine*	2.63 \pm 0.22 ^a	2.16 \pm 0.17 ^{ab}	2.08 \pm 0.1 ^{ab}	2.01 \pm 0.14 ^b	1.96 \pm 0.06 ^b	0.03

** Different letters indicate statistically significant differences between groups ($P < 0.05$)

Table 5. Cecum and fecal parameters according to increasing tannin levels in the diet

	T0 ($\bar{X} \pm S_x$)	T1 ($\bar{X} \pm S_x$)	T2 ($\bar{X} \pm S_x$)	T3 ($\bar{X} \pm S_x$)	T4 ($\bar{X} \pm S_x$)	P Value
Cecal lactic acid (mmol/l)	2.12 ± 0.93	2.37 ± 1.04	6.69 ± 2.05	5.63 ± 2.60	8.82 ± 1.96	0.07
Cecal pH	6.02 ± 0.22	5.84 ± 0.11	6.17 ± 0.13	6.41 ± 0.20	5.92 ± 0.09	0.63
Cecal NH ₃ (mmol/l)	0.5 ± 0.07	0.17 ± 0.04	0.47 ± 0.15	0.29 ± 0.1	0.27 ± 0.08	0.12
VFA						
Acetic acid (%)	69 ± 1.98	68.3 ± 2.96	71 ± 2.12	69.1 ± 0.96	68.5 ± 1.17	0.89
Propionic acid (%)	10.3 ± 1.17	8.9 ± 0.93	9.9 ± 1.08	9.3 ± 0.78	8.1 ± 0.63	0.46
Butyric acid (%)	19.4 ± 2.02	22.0 ± 2.75	17.9 ± 1.51	20.3 ± 1.66	22.2 ± 0.87	0.51
Others* (%)	1.32 ± 0.29	0.79 ± 0.11	1.23 ± 0.26	1.27 ± 0.21	1.2 ± 0.13	0.42
Total VFA (μmol/g)	164 ± 10.54	179 ± 8.21	182 ± 17.04	161 ± 11.56	188 ± 8.71	0.41
(A+B)/P**	4.41 ± 0.65	3.86 ± 0.61	4.7 ± 0.42	4.07 ± 0.52	3.5 ± 0.19	0.29
A/P	3.84 ± 0.59	3.43 ± 0.57	4.13 ± 0.4	3.57 ± 0.43	3.13 ± 0.19	0.41
Feces						
Fecal DM (%)	59.9 ± 2.49	57.3 ± 1.28	60.3 ± 2.16	57.9 ± 2.84	63.1 ± 2.25	0.42
Fecal score	3.63 ± 0.18	3.75 ± 0.19	3.44 ± 0.27	3.56 ± 0.22	3.88 ± 0.21	0.66
Fecal NH ₃ (mmol/l)	2.67 ± 0.11	5.17 ± 0.26	5.66 ± 0.24	5.13 ± 0.44	2.88 ± 0.44	0.55

* Others: Isobutyric Acid + Valeric acid.

** A: Acetic acid, B: Butyric acid, P: Propionic acid.

Table 6. Fecal *Eimeria* oocyst count, plasma IgA, IgG, IgE levels, and mortality rate

	T0 ($\bar{X} \pm S_x$)	T1 ($\bar{X} \pm S_x$)	T2 ($\bar{X} \pm S_x$)	T3 ($\bar{X} \pm S_x$)	T4 ($\bar{X} \pm S_x$)	P Value
<i>Eimeria</i> oocyst count*	41667 ± 15937 ^a	13567 ± 4318 ^{ab}	13958 ± 8356 ^{ab}	3317 ± 943 ^{bc}	1600 ± 582 ^c	0.005
IG						
IgA (g/L)	0.268 ± 0.016	0.248 ± 0.051	0.236 ± 0.041	0.201 ± 0.059	0.283 ± 0.034	0.703
IgG (g/L)	0.241 ± 0.013	0.235 ± 0.007	0.253 ± 0.008	0.231 ± 0.015	0.237 ± 0.008	0.635
IgE (IU/ml)	23.4 ± 1.118	25.4 ± 1.518	21.9 ± 0.89	24.2 ± 1.205	23.1 ± 0.573	0.265
Mortality rate*	13.89 ^b	9.72 ^{ab}	16.67 ^b	16.67 ^b	2.78 ^a	0.047

* Different letters indicate statistically significant differences between groups ($P < 0.05$).

DISCUSSION

In this study, it was observed that even when the tannin level in the diet was increased to 2%, there was no significant effect on body weight and feed intake parameters, either during the starter or grower phases (Table 2). Previous studies have also reported that adding 2% quebracho condensed tannin to pheasant diets (Marzoni et al., 2005b), 6% quebracho condensed tannin to partridge diets (Liukkonen-Anttila et al., 2001), 1.5% and 2.5% quebracho condensed tannin to duck diets (Marzoni et al., 2020), and 0.25% chestnut tannin to broiler diets (Schiavone et al., 2008) did not alter feed intake or body weight. Studies reporting that tannins suppress growth performance are mostly related to faster-growing broilers (Hassan et al., 2003; Buyse et al., 2021; Pertiwi et al., 2022). It seems that tannins do not affect the growth performance of relatively slower-growing birds such as partridges, pheasants, and ducks, and the outcomes may vary depending on the chemical structure and level of tannin added. It is possible that birds like partridges and pheasants, whose digestive systems and metabolism are adapted to the natural environment, are less affected by consuming tannin-rich feed materials in intensive farming conditions, with respect to parameters such as feed intake, feed efficiency, and growth performance.

The study found that increasing doses of tannins did not change the relative weights of the carcass, liver, gizzard, stomach, and intestines (Table 4). In agreement with this study, tannins added to the diets of partridges did not affect carcass, gizzard, or liver weights (Liukkonen-Anttila et al., 2001), and similar results were observed in broilers (Ahmed et al., 1991; Xi et al., 2022; Marzoni et al., 2020) and ducks (Marzoni et al., 2005a). In this study, the weight of the large intestine in the control group was statistically higher than in the 1% and 2% tannin groups, and numerically higher than in the 0.25% and 0.5% tannin groups. Consistent with these findings, a study by Wang et al. (2020) in pigs reported that tannin reduced the weight of the large intestine. Another study in broilers (Perić et al., 2022) reported that tannin addition to the diet numerically reduced the thickness of the tunica

muscularis in the large intestine. It is suggested that the reduction in the weight of the large intestine observed in this study might be due to a decrease in the thickness of the tunica muscularis. It is known that one of the most significant effects of adding antibiotics to feed is reducing gut thickness to increase nutrient absorption (Miyakawa et al., 2024). In this study, it is hypothesized that tannins may have prevented an increase in gut thickness by reducing the number of pathogenic bacteria and protozoan coccidiosis agents, or by increasing butyric acid production, thus preventing pathogen colonization. The observed increase in lactic acid levels in response to higher tannin levels also supports this view. Furthermore, the anti-inflammatory effects of tannins have been documented in previous studies (Huang et al., 2018). The increased tannin intake may have mitigated the effects of a potential colonic inflammation, preventing the thickening of the intestinal tissue and thereby preventing an increase in the weight of the large intestine. The findings of Redondo et al. (2022), which reported that adding tannins to broiler feed improved intestinal lesion scores, further support this hypothesis.

This study determined that the addition of tannins to partridge chick feed did not alter the digestibility of dry matter, organic matter, or crude fat. Consistent with these findings, studies on broilers have reported that tannins added to the diet did not affect the digestibility of dry matter, organic matter, or crude fat (Schiavone et al., 2008; Liu et al., 2020; Kumar et al., 2007; Rezar and Salobir, 2014). The presence of polyphenolic compounds in the diet can have negative consequences, such as reducing the effectiveness of digestive enzymes and decreasing the digestibility of nutrients, particularly proteins and amino acids (Butler and Rogler, 1992). Although differences in crude protein digestibility among the groups were statistically insignificant in this study, it was observed that tannins in the diet tended to reduce crude protein digestibility. Some studies report tannins do not affect protein digestibility (Schiavone et al., 2008; Chang and Fuller, 1964; Viveros et al., 2011), while others report that tannins increase protein digestibility (Liu et al., 2020; Brufau et al., 1998; Godoy et al., 2024).

In the present study, it was observed that tannin at a level of 2% in partridge chick feed significantly reduced calcium digestibility, while lower levels of tannin only resulted in a numerical reduction. Supporting these findings, it has been reported that high-tannin sorghum in broiler (Hassan et al., 2003) and rabbit (Al-Mamary et al., 2001) diets decreased calcium absorption. A study on rats also reported that tannic acid reduced calcium digestibility (Chang et al., 1994). Contrary to the results of this study, Kumar et al. (2007) found that calcium digestibility was not reduced in chickens fed with high-tannin red sorghum. Furthermore, it has been shown that the addition of hydrolyzable chestnut tannins at levels of 0.07% and 0.2% did not affect calcium absorption in broiler diets (Rezar and Salobir, 2014). This study found that tannin levels up to 2% in the diet did not affect phosphorus and magnesium digestibility, but these were numerically lower in the treatment groups compared to the control group. Similarly, the addition of 0.07% and 0.2% chestnut tannins to broiler diets was reported to not affect on phosphorus absorption (Rezar and Salobir, 2014). Additionally, it has been shown that high-tannin sorghum does not alter phosphorus and magnesium absorption in broilers (Kumar et al., 2007). High levels of sorghum tannins in rabbit diets (Al-Mamary et al., 2001) and tannic acid in rats (Chang et al., 1994) have been reported not to affect magnesium absorption. However, Hassan et al. (2003) reported that high doses of tannins reduced magnesium and phosphorus digestibility in broilers. The finding that condensed tannin reduced calcium digestibility while not affecting magnesium and phosphorus digestibility suggests that tannins may preferentially bind to calcium compared to magnesium and phosphorus.

It was observed that the number of *Eimeria* oocysts in the feces decreased with increasing levels of tannins. Studies in broilers have reported that tannic acid (Tonda et al., 2018; Choi et al., 2022), quebracho condensed tannin (Marzoni et al., 2020), and gall oak (*Quercus infectoria*) (Ghafouri et al., 2023) significantly reduced the number of *Eimeria* oocysts. In contrast, some studies have found that tannic acid (Mansoori and Modirsanei, 2012)

and quebracho condensed tannin (McCann et al., 2006) did not affect the number of oocysts. While the majority of studies in poultry align with the findings of this study, it is thought that the effect of tannins on *Eimeria* spp. may vary depending on the source, chemical structure, and level of tannin.

The study found that increasing doses of tannins in partridge chick diets did not affect serum IgA, IgG, or IgE levels (Table 6). While some studies report that tannins do not affect plasma immunoglobulin levels (Liu et al., 2020; Xu et al., 2023), there are also studies suggesting that tannins suppress the immune system, reducing IgG and IgM levels (Marzo et al., 1990). A review of previous studies indicates that the effects of tannins on the immune system vary significantly depending on the chemical structure, dose, and experimental conditions. Most studies have been conducted by experimentally infecting with pathogens. Detailed studies are needed to better understand the effects of tannins on immune proteins.

The study suggests that the level of condensed tannin consumption affects survival rates. In this study, the survival rate of the group receiving 2% tannin was higher than that of the other groups. It was observed that the survival rates of groups receiving lower levels of tannin were similar to those of the control group. Studies on pheasants (Marzoni et al., 2005b) and broilers (Redondo et al., 2022) reported that quebracho condensed tannin did not affect mortality. The improved survival rate observed in this study is thought to be due to the antimicrobial, antiparasitic, and antioxidant properties of tannins. The improvement in survival rates in the current study is thought to be due to the antimicrobial, and antiparasitic effects, and improvements in antioxidant capacity, which positively impact gut health. *Eimeria* spp. settles in the mucosal epithelium of the digestive tract and proliferates. It disrupts the integrity of the mucosa in the digestive tract, triggering intestinal inflammation, haemorrhage, and diarrhea, which leads to productivity loss and death in poultry (Choi and Kim, 2020). It is an accepted fact that tannins have anticoccidial effects. Tannins stimulate the immune system in chickens by

forming complexes with parasitic enzymes and metal ions required for *Eimeria* spp. (Chung et al., 1998; Min and Hart, 2003). The significant decrease in the number of *Eimeria* oocysts with increasing tannin levels in the current study supports the view that this may have contributed to increased survival rates. Additionally, the tendency of lactic acid to increase in the cecum suggests the possibility of an increased number of lactic acid bacteria. Lactic acid bacteria are a major probiotic source that regulates gut health and has immunomodulatory, anti-inflammatory, and antimicrobial effects. They are known to inhibit the growth of pathogenic bacteria by producing bacteriocins and short-chain fatty acids (Sirisopapong et al., 2023). It is believed that the positive effects of the increased lactic acid concentration, along with the increased tannin levels, may have contributed to the reduction in mortality rates. Furthermore, it can be said that the anti-inflammatory effects of tannins (Huang et al., 2018) and their effect in reducing intestinal lesions (Xu et al., 2023) may have prevented an increase in large intestine weight and contributed to the reduction in mortality by improving intestinal health.

In the study, it was observed that the addition of condensed tannins to the diets of partridge chicks did not change the cecal NH_3 concentration, but the treatment groups were slightly lower than the control group (Table 5). It has been reported that tannic acid added to pig diets reduces cecal NH_3 concentration (Biagi et al., 2010). Tannins have been reported to reduce bacterial proteolytic activity in the cecum, thereby decreasing ammonia release (Biagi et al., 2010). In the present study, it can be suggested that the numerical decrease in NH_3 concentration in the cecum is due to the reduction in bacterial protein degradation by the tannin used. The possibility of a low large intestine volume and a high transit rate in partridges may lead to the opinion that the low fermentation rate also contributed to these results. Although the differences in cecal lactic acid content among the groups were found to be statistically insignificant, it was observed that the tannin added to the diet tended to increase cecal lactic acid levels (Table 5). It has been reported that hydrolyzable tannins given to pigs increase

the number of lactic acid bacteria in the intestines (Brus et al., 2013). Moreover, it is known that tannins have effects on digestive enzymes (Strumeyer and Malin, 1970; Butler and Rogler, 1992). It is considered that the digestion of some easily degradable carbohydrates may have been altered by affecting enzymatic digestion in the cecum. While lactic acid increased, no change was observed in cecal pH (Table 5). It can be assumed that the lactic acid produced was rapidly fermented and absorbed, causing no change in cecal pH levels. Other studies have reported that tannic acid did not reduce intestinal pH in pigs (Wang et al., 2020), whereas it lowered pH in broilers (Mašek et al., 2014).

The total VFA (volatile fatty acids) amount was numerically higher in the 0.25%, 0.5%, and 2% tannin groups compared to the control group, but the proportions of acetic acid, propionic acid, butyric acid, and other VFAs were found to be similar (Table 5). In line with the present study, it has been reported that tannic acid and gallic acid added to broiler diets (Mašek et al., 2014) and condensed tannins (Cross et al., 2011) increased the total VFA amount without changing the proportions of acetic, butyric, and propionic acids. Additionally, tannic acid has been reported to increase the total VFA amount in rats (Barszcz et al., 2011). In contrast to these reports, another study reported that tannic acid added to broiler diets tended to decrease cecal VFA production (Choi et al., 2022). It is known that tannin supplementation reduces digestion and nutrient absorption in the small intestine (Barszcz et al., 2011). In the present study, it is thought that due to this effect of tannins, the increased amount of nutrients passing undigested into the cecum may have numerically increased cecal fermentation. Additionally, it can be stated that tannins are resistant to animal digestive enzymes but can be broken down by bacterial degradation (Bhat et al., 1998). In the present study, it is considered that the undigested nutrients reaching the cecum due to the effects of tannins might have been used as a substrate for microbial fermentation, and the slight increase in volatile fatty acids might be due to this reason. Furthermore, it is known that tannins suppress intestinal pathogens and affect gut microbiota (Elizondo

et al., 2010; Costabile et al., 2011; Daing et al., 2017; Reyes et al., 2017). Thanks to this property, it is predicted that the tannins added to the diet suppressed intestinal pathogens and improved cecal fermentation.

In the present study, it was observed that the addition of tannin up to 2% in the diets of partridge chicks did not affect fecal dry matter, fecal score, and fecal ammonia levels, but the 2% tannin group tended to increase fecal score and dry matter. In line with these results, it has been shown that the addition of 2.5% tannin to duck diets (Marzoni et al., 2005a) and the addition of 2% tannin to pheasant diets (Marzoni et al., 2005b) did not affect fecal dry matter. Contrary to the findings, another study conducted in chickens (Marzoni et al., 2020) reported that the addition of 2% quebracho condensed tannin to the diet, as well as 0.07% and 0.2% hydrolyzable tannin (Rezar and Salobir, 2014), significantly increased fecal dry matter.

In the present study, it was observed that quebracho condensed tannin did not change fecal ammonia levels, similar to cecal ammonia levels. In line with the findings, it has been reported that high doses (6%) of quebracho condensed tannin did not affect fecal ammonia levels in partridges (Liukkonen-Anttila et al., 2001), and tannins obtained from cowpea and tea did not affect fecal ammonia levels in rats (Chang et al., 1994). Schiavone et al. (2008) showed that quebracho condensed tannin increased ammonia levels at doses of 0.15% and 0.20% but did not affect it at a dose of 0.25%. In a study conducted on ruminants, it was reported that chestnut tannins reduced fecal ammonia levels (Śliwiński et al., 2004). The different results obtained in studies regarding ammonia levels indicate that factors other than tannins may also affect ammonia production.

CONCLUSION

The study has determined that the addition of condensed tannins to the diet of partridge chicks does not negatively affect growth performance but does improve survival rates. Considering that partridges are primarily produced for hobby and hunting purposes rather than

for meat production, it can be concluded that adding 2% condensed tannins to the diet of partridge chicks can enhance survival rates and protect against coccidiosis.

ACKNOWLEDGEMENTS

This research project was funded by the Turkish Ministry of Agriculture and Forestry with the project number TAGEM/HSGYAD/A/23/A3/P1/6016.

REFERENCES

- Akkılıç, M., Sürmen, S. (1979) Yem Maddeleri ve Hayvan Besleme Laboratuvar Kitabı. Ankara Üniversitesi Basımevi, Ankara.
- Al-Mamary, M., Al-Habori, M., Al-Aghbari, A., Al-Obeidi, A. (2001) Effects of dietary sorghum tannins on rabbit digestive enzymes and mineral absorption. *Nutrition Research*, 21 (10), 1393-1401. DOI: [https://doi.org/10.1016/S0271-5317\(01\)00334-7](https://doi.org/10.1016/S0271-5317(01)00334-7)
- AOAC (2003) Official methods of analysis of AOAC International, 17th Ed. 2nd Revision. Association of Analytical Communities. Gaithersburg, MD, USA.
- Barszcz, M., Taciak, M., Skomial, J. (2011) A dose-response effects of tannic acid and protein on growth performance, caecal fermentation, colon morphology, and β -glucuronidase activity of rats. *Journal of Animal and Feed Sciences*, 20 (4), 613-625. DOI: <https://doi.org/10.22358/jafs/66219/2011>
- Bhat, T. K., Singh, B., Sharma, O. P. (1998) Microbial degradation of tannins--a current perspective. *Biodegradation*, 9 (5), 343-357. DOI: <https://doi.org/10.1023/a:1008397506963>
- Biagi, G., Cipollini, I., Paulicks, B. R., Roth, F. X. (2010) Effect of tannins on growth performance and intestinal ecosystem in weaned piglets. *Archives of Animal Nutrition*, 64 (2), 121-135. DOI: <https://doi.org/10.1080/17450390903461584>
- Borshchevskaya, L. N., Gordeeva, T. L., Kalinina, A. N., Sineokii, S. P. (2016) Spectrophotometric determination of lactic acid. *Journal of Analytical Chemistry*, 71 (8), 755-758. DOI: <https://doi.org/10.1134/S1061934816080037>
- Brufau, J., Boros, D., Marquardt, R. R. (1998) Influence of growing season, tannin content and autoclave treatment on the nutritive value of near-isogenic lines of faba beans (*Vicia faba* L.) when fed to leghorn chicks. *British Poultry Science*, 39 (1), 97-105. DOI: <https://doi.org/10.1080/00071669889457>
- Brus, M., Dolinšek, J., Cencič, A., Škorjanc, D. (2013) Effect of chestnut (*Castanea sativa* Mill.) wood tannins and organic acids on growth performance and faecal microbiota of pigs from 23 to 127 days of age. *Bulgarian Journal of Agricultural Science*, 19 (4), 841-847.
- Butler, L. G., Rogler, J. C. (1992) Biochemical mechanisms of the antinutritional effects of tannins. In: ACS Publications.
- Buyse, K., Delezie, E., Goethals, L., Van Noten, N., Ducatelle, R., Janssens, G. P. J., Lourenco, M. (2021) Chestnut tannins in broiler diets: performance, nutrient digestibility, and meat quality. *Poultry Science*, 100 (12), 101479. DOI: <https://doi.org/10.1016/j.psj.2021.101479>
- Castillo, A., Schiavone, A., Cappai, M. G., Nery, J., Gariglio, M., Sartore, S., Franzoni, A., Marzoni, M. (2020) Performance of Slow-Growing Male Muscovy Ducks Exposed to Different Dietary Levels of Quebracho Tannin. *Animals (Basel)*, 10(6), 979. DOI: <https://doi.org/10.3390/ani10060979>

- Chandrawathani, P., Premaalatha, B., Jamnah, O., Priscilla, F., Erwanas, A., Lily Rozita, M., Jackie, P., Josephin, S. (2015) Mc Master method of worm egg count from faecal samples of goats: A comparison of single and double chamber enumeration of worm eggs. *Malaysian Journal of Veterinary Research*, 6(1), 81-87.
- Chang, M. C., Bailey, J. W., Collins, J. L. (1994) Dietary tannins from cowpeas and tea transiently alter apparent calcium absorption but not absorption and utilization of protein in rats. *Journal of Nutrition*, 124 (2), 283-288. DOI: <https://doi.org/10.1093/jn/124.2.283>
- Chang, S. I., Fuller, H. (1964) Effect of tannin content of grain sorghums on their feeding value for growing chicks. *Poultry Science*, 43(1), 30-36.
- Choi, J., Kim, W. K. (2020) Dietary application of tannins as a potential mitigation strategy for current challenges in poultry production: A review. *Animals*, 10 (12), 2389. DOI: <https://doi.org/10.3390/ani10122389>
- Choi, J., Tompkins, Y. H., Teng, P.-Y., Gogal Jr, R. M., Kim, W. K. (2022) Effects of tannic acid supplementation on growth performance, oocyst shedding, and gut health of in broilers infected with *Eimeria maxima*. *Animals*, 12 (11), 1378. DOI: <https://doi.org/10.3390/ani12111378>
- Choi, J., Yadav, S., Wang, J., Lorentz, B. J., Lourenco, J. M., Callaway, T. R., Kim, W. K. (2022) Effects of supplemental tannic acid on growth performance, gut health, microbiota, and fat accumulation and optimal dosages of tannic acid in broilers. *Frontiers in Physiology*, 13, 912797. DOI: <https://doi.org/10.3389/fphys.2022.912797>
- Chowdhury, S. P., Khanna, S., Verma, S., Tripathi, A. (2004) Molecular diversity of tannic acid degrading bacteria isolated from tannery soil. *Journal of Applied Microbiology*, 97 (6), 1210-1219.
- Chung, K. T., Wong, T. Y., Wei, C. I., Huang, Y. W., Lin, Y. (1998) Tannins and human health: a review. *Critical Reviews in Food Science and Nutrition*, 38 (6), 421-464. DOI: <https://doi.org/10.1080/10408699891274273>
- Costabile, A., Sanghi, S., Martin-Pelaez, S., Mueller-Harvey, I., Gibson, G. R., Rastall, R. A., Klinder, A. (2011) Inhibition of *Salmonella Typhimurium* by tannins *in vitro*. *Journal of Food, Agriculture and Environment*, 9, 119-124.
- Cross, D. E., McDevitt, R., Acamovic, T. (2011) Herbs, thyme essential oil and condensed tannin extracts as dietary supplements for broilers, and their effects on performance, digestibility, volatile fatty acids and organoleptic properties. *British Poultry Science*, 52 (2), 227-237.
- Daing, I., Pathak, A., Bhat, A., Zargar, A. (2017) Antioxidant and antibacterial potential of condensed tannins containing tree leaves extract. *Veterinary Practitioner*, 18 (1), 118-121.
- Degen, A. A., Becker, K., Makkar, H. P., Borowy, N. (1995) *Acacia saligna* as a fodder tree for desert livestock and the interaction of its tannins with fibre fractions. *Journal of the Science of Food and Agriculture*, 68 (1), 65-71.
- Diana, T. F., Calderano, A. A., Rostagno, H. S., Marques, M. R. d. L., Tavernari, F. d. C., Veroneze, R., Albino, L. F. T. (2022) Apparent calcium retention and digestibility coefficients of limestone with different particle sizes in laying hens. *Scientia Agricola*, 80, e20210258.
- Elizondo, A. M., Mercado, E. C., Rabinovitz, B. C., Fernandez-Miyakawa, M. E. (2010) Effect of tannins on the *in vitro* growth of *Clostridium perfringens*. *Veterinary microbiology*, 145(3-4), 308-314.
- Garcia, R. G., Mendes, A. A., Sartori, J. R., Paz, I. d. L. A., Takahashi, S., Pelícia, K., Komiya, C., Quinteiro, R. (2004) Digestibility of feeds containing sorghum, with and without tannin, for broiler chickens submitted to three room temperatures. *Brazilian Journal of Poultry Science*, 6, 55-60.
- Ghafouri, S. A., Ghaniei, A., Sadr, S., Amiri, A. A., Tavanaee Tamannaee, A. E., Charbgoon, A., Ghiassi, S., Dianat, B. (2023) Anticoccidial effects of tannin-based herbal formulation (*Artemisia annua*, *Quercus infectoria*, and *Allium sativum*) against coccidiosis in broilers. *Journal of Parasitic Diseases*, 47 (4), 820-828.
- Godoy, G. L., Rodrigues, B. N., Agilar, J. C., Biselo, V., Brutti, D. D., Maysonave, G. S., Stefanello, C. (2024) Effects of *Acacia mearnsii* tannins on growth performance, footpad dermatitis, nutrient digestibility, intestinal permeability, and meat quality of broiler chickens. *Animal Feed Science and Technology*, 308, 115875. DOI: <https://doi.org/10.1016/j.anifeedsci.2024.115875>
- Gülşen, N., Umucallılar, H., Kırıkçı, K., Hayırlı, A., Aktümsek, A., Alaşahan, S. (2010) Sunflower oil supplementation alters meat quality but not performance of growing partridges (*Alectoris chukar*). *Journal of Animal Physiology and Animal Nutrition*, 94 (2), 196-203.
- Hassan, I., Elzubeir, E., El Tinay, A. (2003) Growth and apparent absorption of minerals in broiler chicks fed diets with low or high tannin contents. *Tropical animal health and production*, 35, 189-196.
- Huang, Q., Liu, X., Zhao, G., Hu, T., Wang, Y. (2018) Potential and challenges of tannins as an alternative to in-feed antibiotics for farm animal production. *Animal Nutrition*, 4 (2), 137-150. DOI: <https://doi.org/10.1016/j.aninu.2017.09.004>
- Karadas, F., Moller, A. P., Karageçli, M. R. (2017) A comparison of fat-soluble antioxidants in wild and farm-reared chukar partridges (*Alectoris chukar*). *Comparative Biochemistry and Physiology. Part A: Molecular and Integrative Physiology*, 208, 89-94. DOI: <https://doi.org/10.1016/j.cbpa.2017.03.015>
- Kokoszynski, D., Bernacki, Z., Korytkowska, H., Wilkanowska, A., Frieske, A. (2013) Carcass composition and meat quality of Grey Partridge (*Perdix perdix* L.). *Journal of Central European Agriculture*, 14 (1), 0-0.
- Kumar, V., Elangovan, A. V., Mandal, A. B., Tyagi, P. K., Bhanja, S. K., Dash, B. B. (2007) Effects of feeding raw or reconstituted high tannin red sorghum on nutrient utilisation and certain welfare parameters of broiler chickens. *British Poultry Science*, 48 (2), 198-204. DOI: <https://doi.org/10.1080/00071660701251089>
- Liu, H. S., Mahfuz, S. U., Wu, D., Shang, Q. H., Piao, X. S. (2020) Effect of chestnut wood extract on performance, meat quality, antioxidant status, immune function, and cholesterol metabolism in broilers. *Poultry Science*, 99 (9), 4488-4495. DOI: <https://doi.org/10.1016/j.psj.2020.05.053>
- Liukkonen-Anttila, T., Kentala, A., Hissa, R. (2001) Tannins--a dietary problem for hand-reared grey partridge *Perdix perdix* after release? *Comparative Biochemistry and Physiology: Toxicology & Pharmacology*, 130 (2), 237-248. DOI: [https://doi.org/10.1016/s1532-0456\(01\)00244-7](https://doi.org/10.1016/s1532-0456(01)00244-7)
- Mansoori, B., Modirsanei, M. (2012) Effects of dietary tannic acid and vaccination on the course of coccidiosis in experimentally challenged broiler chicken. *Veterinary Parasitology*, 187 (1-2), 119-122. DOI: <https://doi.org/10.1016/j.vetpar.2011.12.016>
- Marzo, F., Tosar, A., Santidrian, S. (1990) Effect of tannic acid on the immune response of growing chickens. *Journal of Animal Science*, 68 (10), 3306-3312. DOI: <https://doi.org/10.2527/1990.68103306x>
- Marzoni, M., Castillo, A., Romboli, I. (2005a) Dietary inclusion of Quebracho (*Schinopsis lorentzii*) tannins on productive performances of growing pheasant females. *Italian Journal of Animal Science*, 4 (Suppl. 2), 507-509.
- Marzoni, M., Castillo, A., Romboli, I. (2005b) Performances of growing Muscovy ducks fed on diets supplemented with Quebracho tannin powder. *Proceedings of the XVII the European Symposium on the Quality of Poultry Meat*, Doorwerth, The Netherlands,

- Marzoni, M., Castillo, A., Franzoni, A., Nery, J., Fortina, R., Romboli, I. (2020) Effects of dietary quebracho tannin on performance traits and parasite load in an Italian slow-growing chicken (White Livorno breed). *Animals*, 10, 1-11.
- Mašek, T., Starčević, K., Mikulec, Ž. (2014) The influence of the addition of thymol, tannic acid or gallic acid to broiler diet on growth performance, serum malondialdehyde value and cecal fermentation. *European Poultry Science/Archiv für Geflügelkunde*, 78.
- McCann, M., Newell, E., Preston, C., Forbes, K. (2006) The use of mannan-oligosaccharides and/or tannin in broiler diets. *International Journal of Poultry Science*, 5 (9), 873-879.
- Min, B., Hart, S. (2003) Tannins for suppression of internal parasites. *Journal of Animal Science*, 81 (14_suppl_2), E102-E109.
- Miyakawa, M. E. F., Casanova, N. A., Kogut, M. H. (2024) How did antibiotic growth promoters increase growth and feed efficiency in poultry? *Poultry Science*, 103 (2), 103278.
- Mossa, A. H., Swelam, E. S., Mohafrash, S. M. M. (2015) Sub-chronic exposure to fipronil induced oxidative stress, biochemical and histopathological changes in the liver and kidney of male albino rats. *Toxicol Rep*, 2, 775-784.
DOI: <https://doi.org/10.1016/j.toxrep.2015.02.009>
- Panayides, P., Guerrini, M., Barbanera, F. (2011) Conservation genetics and management of the chukar Partridge *Alectoris chukar* in Cyprus and the Middle East. *Sandgrouse*, 33, 34-43.
- Perić, L., Žikić, D., Stojčić, M. Đ., Tomović, V., Leskovec, J., Levart, A., Salobir, J., Kanački, Z., Rezar, V. (2022) Effect of Chestnut Tannins and Vitamin E Supplementation to Linseed Oil-Enriched Diets on Growth Performance, Meat Quality, and Intestinal Morphology of Broiler Chickens. *Agriculture*, 12 (11), 1772.
- Pertiwi, H., Rochmi, S. E., Chwen, L. T. (2022) Detrimental Effect of Tannin on Growth Performance, Visceras Weight and Blood Biochemistry in Broiler Chickens Reared Under Tropical Area. *Archives of Razi Institute*.
- Redondo, E. A., Redondo, L. M., Bruzzone, O. A., Diaz-Carrasco, J. M., Cabral, C., Garces, V. M., Lineiro, M. M., Fernandez-Miyakawa, M. E. (2022) Effects of a blend of chestnut and quebracho tannins on gut health and performance of broiler chickens. *PLoS One*, 17 (1), e0254679. DOI: <https://doi.org/10.1371/journal.pone.0254679>
- Redondo, L. M., Chacana, P. A., Dominguez, J. E., Fernandez Miyakawa, M. E. (2014) Perspectives in the use of tannins as alternative to antimicrobial growth promoter factors in poultry. *Frontiers in Microbiology*, 5, 118.
- Reyes, A. W. B., Hong, T. G., Hop, H. T., Arayan, L. T., Huy, T. X. N., Min, W., Lee, H. J., Lee, K. S., Kim, S. (2017) The *in vitro* and *in vivo* protective effects of tannin derivatives against *Salmonella enterica* serovar Typhimurium infection. *Microbial Pathogenesis*, 109, 86-93. DOI: <https://doi.org/10.1016/j.micpath.2017.05.034>
- Rezar, V., Salobir, J. (2014) Effects of tannin-rich sweet chestnut (*Castanea sativa* mill.) wood extract supplementation on nutrient utilisation and excreta dry matter content in broiler chickens. *European Poultry Science/Archiv für Geflügelkunde*, 78.
- Scalbert, A. (1991) Antimicrobial properties of tannins. *Phytochemistry*, 30 (12), 3875-3883.
- Schiavone, A., Guo, K., Tassone, S., Gasco, L., Hernandez, E., Denti, R., Zoccarato, I. (2008) Effects of a natural extract of chestnut wood on digestibility, performance traits, and nitrogen balance of broiler chicks. *Poultry Science*, 87 (3), 521-527.
DOI: <https://doi.org/10.3382/ps.2007-00113>
- Schofield, P., Mbugua, D., Pell, A. (2001) Analysis of condensed tannins: a review. *Animal Feed Science and Technology*, 91 (1-2), 21-40.
- Scholten, M. T., De Boer, I., Gremmen, B., Lokhorst, C. (2013) Livestock farming with care: towards sustainable production of animal-source food. *NJAS: Wageningen Journal of Life Sciences*, 66 (1), 3-5.
- Sigma-Aldrich, C. (1998) Analyzing fatty acids by packed column gas chromatography. Bellefonte: SUPELCO.
- Sirisopapong, M., Shimosato, T., Okrathok, S., Khempaka, S. (2023) Assessment of lactic acid bacteria isolated from the chicken digestive tract for potential use as poultry probiotics. *Animal Bioscience*, 36 (8), 1209.
- Śliwiński, B., Kreuzer, M., Sutter, F., Machmüller, A., Wettstein, H. (2004) Performance, body nitrogen conversion and nitrogen emission from manure of dairy cows fed diets supplemented with different plant extracts. *Journal of Animal and Feed Sciences*, 13 (1), 73-91.
- Soltanpour, P. N., Johnson, G. W., Workman, S. M., Jones Jr, J. B., Miller, R. O. (1996) Inductively coupled plasma emission spectrometry and inductively coupled plasma-mass spectrometry. *Methods of Soil Analysis: Part 3 Chemical Methods*, 5, 91-139.
- Strumeyer, D. H., Malin, M. J. (1970) Resistance of extracellular yeast invertase and other glycoproteins to denaturation by tannins. *Biochemical Journal*, 118(5), 899-900. DOI: <https://doi.org/10.1042/bj1180899>
- Swanson, K. S., Grieshop, C. M., Flickinger, E. A., Bauer, L. L., Healy, H. P., Dawson, K. A., Merchen, N. R., Fahey, G. C., Jr. (2002) Supplemental fructooligosaccharides and mannanoligosaccharides influence immune function, ileal and total tract nutrient digestibilities, microbial populations and concentrations of protein catabolites in the large bowel of dogs. *Journal of Nutrition*, 132 (5), 980-989. DOI: <https://doi.org/10.1093/jn/132.5.980>
- Tietz, N. W. (1995) Clinical guide to laboratory tests. In *Clinical guide to laboratory tests*, pp. 1096-1096.
- Tonda, R. M., Rubach, J. K., Lumpkins, B. S., Mathis, G. F., Poss, M. J. (2018) Effects of tannic acid extract on performance and intestinal health of broiler chickens following coccidiosis vaccination and/or a mixed-species *Eimeria* challenge. *Poultry Science*, 97 (9), 3031-3042. DOI: <https://doi.org/10.3382/ps/pey158>
- Van Soest, P. V., Robertson, J. B., Lewis, B. A. (1991) Methods for dietary fiber, neutral detergent fiber, and nonstarch polysaccharides in relation to animal nutrition. *Journal of Dairy Science*, 74 (10), 3583-3597. DOI: [https://doi.org/10.3168/jds.S0022-0302\(91\)78551-2](https://doi.org/10.3168/jds.S0022-0302(91)78551-2)
- Viveros, A., Chamorro, S., Pizarro, M., Arija, I., Centeno, C., Brenes, A. (2011) Effects of dietary polyphenol-rich grape products on intestinal microflora and gut morphology in broiler chicks. *Poultry Science*, 90 (3), 566-578.
DOI: <https://doi.org/10.3382/ps.2010-00889>
- Wang, M., Huang, H., Hu, Y., Huang, J., Yang, H., Wang, L., Chen, S., Chen, C., He, S. (2020) Effects of dietary microencapsulated tannic acid supplementation on the growth performance, intestinal morphology, and intestinal microbiota in weaning piglets. *Journal of Animal Science*, 98 (5), skaa112.
DOI: <https://doi.org/10.1093/jas/skaa112>
- Xi, Y., Chen, J., Guo, S., Wang, S., Liu, Z., Zheng, L., Qi, Y., Xu, P., Li, L., Zhang, Z., Ding, B. (2022) Effects of tannic acid on growth performance, relative organ weight, antioxidant status, and intestinal histomorphology in broilers exposed to aflatoxin B(1). *Frontiers in Veterinary Science*, 9, 1037046.
DOI: <https://doi.org/10.3389/fvets.2022.1037046>
- Xu, H., Fu, J., Luo, Y., Li, P., Song, B., Lv, Z., Guo, Y. (2023) Effects of tannic acid on the immunity and intestinal health of broiler chickens with necrotic enteritis infection. *Journal of Animal Science and Biotechnology*, 14 (1), 72.
DOI: <https://doi.org/10.1186/s40104-023-00867-8>