



EFFECT OF DIETARY SUPPLEMENTATION WITH ORANGE PEEL POWDER (CITRUS SINENSIS) ON PRODUCTIVE PERFORMANCE AND SELECTED PHYSIOLOGICAL TRAITS OF BROILER CHICKENS

Majeed Jabbar Shakir

Department of Animal Production, College of Agriculture, Tikrit University, Iraq
majeed.j.s033@tu.edu.iq

Abstract: The objective of this investigation was to determine the effect of graded levels of dried sweet orange (*Citrus sinensis*) peel powder supplementation on growth performance and some physiological and biochemical parameters of broiler chicks. A completely randomized design was used, with 180 one-day-old Ross 308 chicks randomly assigned to four dietary treatments (10 birds/replicate, three replications/treatment). The basal diet was fed to the control group (D1) while the groups D2, D3 and D4 were supplemented with 0.5, 1.0 and 1.5% orange peel powder, respectively. The feeding trial was 42 days. At all times during this period, the following performance measurements were taken and recorded, namely: live body weight, weight gain, feed intake and feed conversion ratio. The blood samples were collected for the estimation of serum total cholesterol, triglycerides, aspartate aminotransferase (AST), and alanine aminotransferase (ALT) at the end of the trial. Data were subjected to one-way analysis of variance and linear and quadratic trend analysis was carried out for the increase in inclusion levels. Statistically significant difference was set at $P < 0.05$. The hypothesis in the study was that the bioactive compounds in orange peel such as flavonoids, polyphenols, pectin, and essential oils, may help to enhance nutrient utilization, control lipid metabolism, and preserve the physiological status of liver in broilers.

Keywords: broiler chickens, *Citrus sinensis*, orange peel powder, growth performance, cholesterol, triglycerides, AST, ALT.

Introduction

Recent concerns about efficient and environmentally-friendly poultry production have sparked significant interest regarding the use of plant-based feed additives and agro-industrial by-products as functional ingredients in poultry feeds. In broiler production, these materials are of special interest because they might have not only nutrients, but also biologically active compounds which can modulate digestion, antioxidant defense, lipid metabolism and overall physiological status. In this regard, citrus-processing by-products have gained more and more interest, and considerable amounts of peel and pulp residues are produced during juice processing, thus offering a possibility to utilize the agricultural residues as potentially useful ingredients in poultry feed (Abbasi et al., 2015).

Sweet orange (*Citrus sinensis*) peel is one of the more promising by-products as it is rich in citrus flavonoids, phenolic compounds and essential oils and dietary fiber. The antioxidant and lipid-modulating properties have been studied extensively for some of the major citrus flavanones, such as hesperidin, naringin, hesperetin, and naringenin. The citrus flavonoids have been reported to act on lipid metabolism in multiple ways, such as regulating the synthesis of fatty acids in hepatic cells, stimulating the oxidation of fatty acids and modulating cholesterol and lipoprotein metabolism (Mulvihill et al., 2016; Mahfuz et al., 2022).

Poultry studies have shown that citrus ingredients can have an effect on both productive and physiological responses. Abbasi et al. (2015) tested different concentrations of dried sweet orange pulp in broiler diets and found that it affected various performance and metabolic indicators. They found that dried citrus residues could be added to a broiler ration and that increased levels of dried citrus residues were correlated with decreased levels of liver fat, abdominal fat, and serum triglycerides. Based on these observations, it can be inferred that the use of feed ingredients derived from oranges can affect the deposition of lipid and lipid metabolites in the blood of broiler chickens.

These physiological effects have been observed with dried sweet orange peel. Alzawqari et al. (2016) conducted the study on the use of dried sweet orange peel in the finisher phase and reported that there was no negative effect on BWG, FIR, FCR and carcass characteristics. But supplementation changed lipid-related variables in the serum and enhanced the total antioxidant status. Specifically, serum triglycerides, low-density lipoprotein and very-low-density lipoprotein were lowered, suggesting that the metabolic effects of orange peel may exist without significant changes in growth rate.

Environmental stress effects have also been determined for citrus peel extracts. Alzawqari et al. (2013) investigated the effect of orange and lemon peel extracts on broilers under high ambient temperature. Feeding the birds supplemented with orange peel extract, no significant effect was seen on weight gain or feed intake or feed conversion ratio, but there was an effect on some of the serum biochemical parameters. It is concluded that the physiological responses to citrus compounds can sometimes not correspond to the productive responses, which reinforces the need to examine both productive and biochemical parameters.

Later studies also showed that sweet orange peel meal was metabolizable. Ogunlade et al. (2025) fed a diet supplemented with sweet orange peel meal (25, 50, 75 g/kg diet) to broiler chickens and concluded that no adverse effects were observed on body-weight gain, feed intake, feed conversion ratio or carcass traits. The results also justify the possibility of using orange peel as a nutritional functional component in the development of broiler feeds.

Thus, the present investigation was conducted to study the effect of graded level of orange peel powder (*Citrus sinensis*) on productive performance parameters like live body weight, live body weight gain, feed intake and feed conversion ratio (FCR), blood physiological and biochemical parameters such as blood total cholesterol, triglycerides, aspartate aminotransferase (AST) and alanine aminotransferase (ALT) of broiler chickens.

Materials and Methods

1. Experimental Birds and Management

A total of 180 day-old Ross 308 broiler chicks with approximately equal initial body weight were used as experimental animals in this study. At the arrival, each chick was weighed and then randomly distributed one to each of four dietary treatments in a completely randomized design. Total birds used per dietary treatment were 15 replicates in each treatment and a total of 3 dietary treatments were performed (45 birds). All birds were reared in the same and similar conditions during the entire experimental period.

Feed and water were supplied ad libitum throughout the whole experimental period. Birds were caged in floor pens equipped with feeders and drinkers and each pen was a replicate in the experiment. The brooding temperature was adjusted in accordance with the Ross 308 temperature management guidelines and gradually decreased with the age of birds. All the experimental groups were given the same treatment for lighting, vaccination and health management.

2. Experimental Design and Dietary Treatments

180 broiler chicks were randomly allocated to four dietary treatments, namely; D1: Basal diet (control group), D2, D3 and D4: Basal diet with 0.5, 1.0 and 1.5% orange peel powder. Experiment was conducted in a completely randomized design (CRD). The replicate pen was considered the experimental unit for the evaluation of productive performance.

3. Preparation of Orange Peel Powder

The fresh sweet orange (*Citrus sinensis*) peels were collected from a single uniform source to reduce the variability of their chemical composition in the batch. Thoroughly cleaned off with clean running water to remove any foreign materials and dirt that might be attached to them, and then chopped into small bits. All pieces were dried in the air-dried in a well-ventilated area away from direct sunlight until a stable constant weight was achieved. Or, the peels could be dried in a forced air oven at about 45–50°C and then ground in an electric mill to form a uniform powder. The powder was then screened and stored in dry and air tight containers up to use in the experimental diets.

4. Experimental Diets

The nutritional requirements of Ross 308 broiler chickens were met with the experimental diets. The feeding program consisted of two phases: a starter diet (0–21 days of age), and a grower/finisher diet (22–35 days of age). Balanced diets were prepared in order to have similar amounts of metabolizable energy, crude protein, essential amino acids, calcium and available phosphorus. The amounts of orange peel powder used were 0%, 0.5%, 1.0% and 1.5% for experimental diets D1, D2, D3 and D4, respectively. All diets were made under the same conditions and fed as a mash. Throughout the experimental period feed and drinking water were allowed ad libitum.

5. Productive Performance Measurements

Growth performance was measured during the entire experimental period by

measuring live body weight, body-weight gain, feed intake and feed conversion ratio (FCR) for each replicate. The body weight of the birds was recorded at the start of the experiment to get initial weight, and then at weekly intervals throughout the trial period until the end, when the body weight was recorded at 35 days old. Average live body weight was then calculated by dividing the total live body weight of the birds by the number of birds, in each replicate, thus, average live body weight (g/bird) = total live body weight of birds/numbers of birds. Body-weight gain (g) of each experimental period was calculated by subtracting the initial body weight from the final body weight of each period and cumulative body-weight gain for the experimental period from 1 to 35 days of age was also calculated by subtracting the initial body weight from the final body weight of the experimental period. Feed intake was determined by carefully weighing the amount of feed offered and the amount of residual feed left in each replicate and was defined as feed offered – feed remaining; the cumulative feed intake was calculated for the entire experimental period. Lastly, FCR was determined as feed intake (g) / body-weight gain (g). Lower FCR value was taken as an evidence of more efficient utilization of feed and it was responsible to the fact that the birds used less feed to gain each unit of weight gain.

6. Blood Collection and Serum Preparation

Blood samples were taken at the end of the experimental period from a representative random sample of birds from each experimental group. In this case, the samples used were 3 birds each from 3 replicates per treatment and 36 birds for all treatments for biochemical analyses. Blood was collected from the brachial (wing) vein of each bird selected and directly placed in plain tubes without anticoagulant, and the total amount of blood collected was about 5 mL. The samples were allowed to clot at room temperature and then centrifuged at c. 3000 rpm for 10-15 min to isolate the serum. The serum was carefully processed and pipetted into clean, labeled eppendorf tubes and stored at –20° until biochemical analyses were performed.

7. Biochemical Analyses

Blood samples were taken from three birds each from experimental groups at the end of experimental period. The samples were left to clot, and then centrifuged to obtain the serum for biochemical evaluation of parameters of lipid profile (total cholesterol and triglycerides) and liver function enzymes (aspartate aminotransferase [AST] and alanine aminotransferase [ALT]). All analysis were carried out by commercially available diagnostic kits, following manufacturers' instructions and protocols.

Serum total cholesterol: The total cholesterol level was determined by the level of serum total cholesterol, measured with a commercial cholesterol diagnostic kit according to the manufacturer's instructions, and expressed in the International units/dL (mg/dL).

Serum triglycerides: The serum triglyceride concentration was measured by an enzymatic colorimetric method and the results were expressed in mg/dl.

Aspartate aminotransferase (AST): Serum AST activity was determined by commercial diagnostic kit, following manufacturer's protocol and the enzyme activity is expressed in Units per liter (U/L).

Alanine aminotransferase (ALT): In blood, ALT activity was determined by the use of a commercial diagnostic kit according to the manufacturer's instructions and presented as U/L.

8. Statistical Analysis

All data obtained were statistically analyzed by completely randomized design

(CRD) and the effect of the dietary treatments was analyzed by one way analysis of variance (ANOVA) for the measured variables. The means were separated in the ANOVA and compared among the different treatments by a Tukey's multiple-range test to determine the differences among the treatments. Furthermore, since orange peel powder was added to the diets in a graded manner, orthogonal polynomial contrasts (linear and quadratic) were used to determine the trend of response to the increasing level of orange peel supplementation – whether the trend was linear or curvilinear as the level increased. All analyses were performed with a probability level of $P < 0.05$ considered significant.

Results

Orange peel powder had a significant impact on broiler body weight during the whole experimental period. After the 1st wk, birds in Group-D2 (0.5% orange peel powder) had the highest body weight (152.66 g) while birds in Group-D3 (0.5% crude orange peel powder) had the lowest body weight (145.99 g). The body weight responses were different in the second and third weeks but, overall, tended to be higher in the supplemented groups than in the control after the fourth week. The body weight of the animals at week 5 showed that Group-D2, D3 and D4 had the highest body weight, 2291.66 g, 2273.33 g and 2253.33 g respectively, while Group-D1 (Control) had the lowest body weight 2212.88 g. The superscript letters denote that D2 and D3 were significantly better than the control at the last measurement while D4 had an intermediate response. The final body weight was 3.56% heavier in D2, 2.73% heavier in D3 and 1.83% heavier in D4 than the control. Therefore, the final body weight was not linearly related to the orange peel powder concentration and the highest numerical response was found for the supplementation level of 0.5% orange peel powder.

Table 1. Effect of Orange Peel Powder (*Citrus sinensis*) in diet on Body Weight of Broiler Chickens

Groups	Age (day)				
	Week-1	Week-2	Week-3	Week-4	Week-5
Control-D1	148.65 ^{ab}	520.88 ^a	1051.10 ^a	1619.55 ^b	2212.88 ^c
Group-D2	152.66 ^a	490.10 ^b	1063.55 ^a	1663.77 ^a	2291.66 ^a
Group-D3	145.99 ^b	527.99 ^a	1017.77 ^c	1661.21 ^b	2273.33 ^a
Group-D4	147.77 ^{ab}	508.99 ^b	1049.11 ^b	1660.32 ^a	2253.33 ^b
<i>P value</i>	*	*	*	*	*

Means within the same row bearing different superscript letters (a, b, c) are significantly different at $P \leq 0.05$, whereas means bearing the same letter are not significantly different. Values are expressed as mean $\pm$ standard error (SE)

Group D2, 3, and 4: add Orange Peel Powder (*Citrus sinensis*) 0.5, 1.0, and 1.5% orange peel powder in diet respectively.

Dietary treatments had a significant effect on body weight gain for weeks 1–4 and the entire experimental period, but not for week 5. Bird fed D2 gained the most

(110.08 g) during the first week and the least was observed in birds fed D3 (104.74 g). D3 had the largest increase (382.00 g) in week 2, and the largest value in week 4 (643.55 g). The highest increase in the D2 was seen in week 3 (573.44 g). After week 5 the weight gain values varied between 586.67 g to 627.89 g among the diet groups and were not significant.

Taken overall from week 1 to 5, D2 had the highest total weight gain (2249.09 g), and D3 the second highest total weight gain (2241.20 g). The cumulative gains were lower in the D4 and the control (D1) group with 2211.10 g and 2169.71 g, respectively. However, total body weight gain was greater in D2, D3 and D4 by 3.66%, 3.29% and 1.91% respectively when compared with the control. Statistically, both D2 and D3 were equal in significance letter and significantly higher than the control; and D4 was intermediate, but not significantly different from the control or the higher groups.

Table 2. Effect of Orange Peel Powder (*Citrus sinensis*) in diet on Body Weight Gain of Broiler Chickens

Age (day)	Groups				P value
	Control D1	Group-D2	Group-D3	Group-D4	
Week-1	106.47 ^b	110.08 ^a	104.74 ^b	107.20 ^{ab}	*
Week-2	371.67 ^a	337.44 ^b	382.00 ^a	361.55 ^{ab}	*
Week-3	529.78 ^{ab}	573.44 ^a	489.78 ^b	534.11 ^{ab}	*
Week-4	568.44 ^b	600.22 ^{ab}	643.55 ^a	621.55 ^{ab}	
Week-5	593.33	627.89	621.11	586.67	n.s
Week-1-5	2169.71 ^b	2249.09 ^a	2241.20 ^a	2211.10 ^{ab}	*

Means within the same row bearing different superscript letters (a, b, c) are significantly different at $P \leq 0.05$, whereas means bearing the same letter are not significantly different. Values are expressed as mean $\pm$ standard error (SE)

Group D2, 3, and 4: add Orange Peel Powder (*Citrus sinensis*) 0.5, 1.0, and 1.5% orange peel powder in diet respectively.

Orange peel powder had relatively limited effects on feed consumption during the early and middle stages of the experiment. No significant differences in feed intake were observed among groups during weeks 1, 2, 3, or 4. A significant effect appeared only during week 5, when D3 and D4 showed higher feed intake than D2 and the control. For the entire experimental period, cumulative feed intake was 3381.10 g in Control-D1, 3491.09 g in D2, 3329.46 g in D3, and 3458.21 g in D4. D2 showed the highest cumulative intake, whereas D3 showed the lowest. The Duncan letters indicate that D2 differed significantly from D3, while the control and D4 occupied an intermediate position and did not differ significantly from either extreme group. Compared with the control, cumulative feed intake increased by about **3.25% in D2**, decreased by **1.53% in D3**, and increased by approximately **2.28% in D4**. The combination of relatively low feed intake and high weight gain in D3 is particularly relevant when interpreting feed efficiency.

Table 3. Effect of Orange Peel Powder (*Citrus sinensis*) in diet on Feed Intake of Broiler Chickens

Age (day)	Groups				P value
	Control D1	Group-D2	Group-D3	Group-D4	
Week-1	122.33	122.00	119.81	121.33	n.s
Week-2	432.66	420.65	449.88	435.22	n.s
Week-3	756.77	803.33	749.88	783.66	n.s
Week-4	1000.44	1004.44	933.44	981.32	n.s
Week-5	1068.89 ^b	1140.66 ^a	1076.44 ^b	1136.66 ^a	*
Week-1-5	3381.10 ^{ab}	3491.09 ^a	3329.46 ^b	3458.21 ^{ab}	*

Means within the same row bearing different superscript letters (a, b, c) are significantly different at $P \leq 0.05$, whereas means bearing the same letter are not significantly different. Values are expressed as mean $\pm$ standard error (SE)

Group D2, 3, and 4: add Orange Peel Powder (*Citrus sinensis*) 0.5, 1.0, and 1.5% orange peel powder in diet respectively.

Feed conversion ratio (FCR) was not significantly different between the dietary treatments for week 1, 2, 3 and 5 of the trial. A statistically significant effect was only seen in week 4. The control group had the poorest feed conversion during this week (1.76) which indicates that they needed more feed to gain the same amount of weight. The D2 and D4 groups, however, had significantly lower FCR values, 1.45 and 1.51, respectively, which signified better feed utilization efficiency. The D3 group, with an FCR of 1.66, was in between these two groups.

The respective FCRs for the control (D1), D2, D3 and D4 were calculated as 1.55, 1.54, 1.48 and 1.52 during the entire experimental period. Groups that received 1.0% orange peel powder (D3) had the lowest FCR value, indicating that their feeds were most effective. It had an FCR of approximately 4.52% lower as compared to the control group. Based on significance letters reported, D3 differed significantly from the control while D2 and D4 were statistically intermediate, not being clearly different from either the control or D3.

Table 4. Effect of Orange Peel Powder (*Citrus sinensis*) in diet on Feed Conversion Ratio of Broiler Chickens

Age (day)	Groups				P value
	Control D1	Group-D2	Group-D3	Group-D4	
Week-1	1.14	1.10	1.14	1.13	n.s
Week-2	1.16	1.24	1.17	1.22	n.s
Week-3	1.43	1.39	1.53	1.41	n.s
Week-4	1.76 ^a	1.66 ^{ab}	1.45 ^b	1.51 ^b	*
Week-5	1.80	1.81	1.72	1.91	n.s
Week-1-5	1.55 ^a	1.54 ^{ab}	1.48 ^b	1.52 ^{ab}	*

Means within the same row bearing different superscript letters (a, b, c) are significantly different at $P \leq 0.05$, whereas means bearing the same letter are not significantly different. Values are expressed as mean $\pm$ standard error (SE)

Group D2, 3, and 4: add Orange Peel Powder (*Citrus sinensis*) 0.5, 1.0, and 1.5% orange peel powder in diet respectively.

From the results of this study, it can be concluded that the supplementation of broiler diet with the addition of orange peel powder could improve growth performance particularly at moderate levels. The significant differences were observed

in the final body weight and cumulative weight gain of the groups receiving 0.5% and 1.0% orange peel powder and the overall feed conversion ratio (FCR), which was most evident in the 1.0% powder group. Important, however, is that the increase in gains did not occur in all instances with an increase in feed intake. The D3 group had a relatively high cumulative weight gain, and numerically the lowest total feed consumption of all groups, leading to the lowest overall FCR for all treatments. This would indicate that the response was likely related to the increased use of the nutrients the birds were eating, as opposed to just eating more. This interpretation aligns with the findings of Abd El Latif et al. (2023) which analyzed different concentrations of orange peel meal used in broiler diets. They found that body weight gain and FCR were significantly affected by orange peel meal especially during grower phase and throughout the experimental period, while feed intake was not significantly influenced by orange peel meal. They had been used at much higher rates than here, but the trend is the same: feed efficiency and growth rate may be improved without an increase in feed intake.

The diversity in phytochemicals in the peel of *Citrus sinensis* is a plausible biological explanation. Citrus derived materials include flavonoids (hesperidin and naringin), pectic compounds, phenolics and volatile. The digestive secretions, nutrient utilisation, intestinal barrier function, oxidative balance and intestinal microbial environment are all reported to be altered by phytochemical feed additives. However, the extent of the effects depends on the botanical origin, processing technique, dose, basal diet, and production conditions (Abdelli et al., 2021).

More specifically, Cisse et al. (2023) described a standardised extract from citrus consisting of hesperidin, naringin, eriocitrin and pectic oligosaccharides and other components. This extract did not significantly affect cecal microbiota but supplementation increased the final broiler growth performance. This is a relevant remark as it suggests that citrus compounds could exert their action through several physiological processes as well as modulation of gut microbiota.

Recent experimental studies also suggest a function for citrus flavonoids in the intestinal function. Liu et al. (2024) showed that dietary hesperidin, especially when combined with other phytochemical compounds, increased average daily gain and final body weight. These changes were correlated with enhanced villus architecture in the intestine, increased barrier function of the tight junctions, modulation of inflammation and beneficial alterations in cecal microbial metabolites. The results presented here provide a plausible mechanism for the improvements in weight gain and feed efficiency seen with the use of whole orange peel powder as opposed to purified hesperidin. Nevertheless, such mechanisms must be considered as hypotheses to be explained by future experimental tests because in this experiment there was no direct measurement of intestinal morphology, digestive enzymes activity and microbiota.

The final weight gain observed in D2 and D3 is consistent with the results obtained with orange residues at relatively low dietary concentrations. In broiler diets, dried sweet orange residues at 4 different levels (0.5, 1.0, 1.5, and 2.0%) were tested by Abbasi et al. (2015), who reported beneficial effect on the performance related traits, but the response varied according to the different traits tested and FCR was not consistently affected. Of special interest is that the inclusion rates were comparable in both of those studies and the present study and that this showed the potential for using orange by-products as functional feed ingredients, and not only as key replacement ingredients in conventional feedstuffs.

Similarly, Vlaicu et al. (2020) found that orange peel supported broiler

bodyweight increase, and correlated with positive changes in the population of intestinal bacteria, such as a decrease in the number of pathogenic microorganisms, and an increase in the quantity of *Lactobacillus* spp. This may reduce the competition for nutrients and create intestinal conditions that will favour better feed utilization. However, as mentioned above, it is not a certainty that microbial modification is all that is needed, as other studies have shown performance improvement from citrus extracts without significant changes in microbiota.

What is noteworthy is the superior FCR of the D3. The 1.0% group achieved a FCR of 1.48 while the control fed 1.55, representing about a 4.5% improvement. Another meta-analysis of the effect of phytogenic in broiler nutrition reported that the additives can positively affect the average daily gain and improve feed conversion, but this effect varies depending on the experimental environment, the level of microbial challenge, and the type of additives and their dosage (Polycarpo et al., 2022). The present data are consistent with this broader trend, the negative impact of the moderate level (1.0%) being greater on feed efficiency than the high level tested.

One of the most important aspects of the results is that there is no simple dose-dependent response. The supplementation level of 1.5% did not result in any further improvement although D2 gave the maximum weight and cumulative gain while D3 gave the best FCR. This is possibly due to the fact that the relatively high amount of structural carbohydrates in the peel may be balanced by the beneficial bioactive components. Fruit peels have antioxidant and antimicrobial properties and can also provide substantial amounts of fibre and some antinutrients that could limit nutrient availability if they are used in excessive amounts. Sugiharto (2023) pointed out that the nutritional content of the fruit-peel products for broilers is highly dependent on the inclusion level and processing of the products, as too high fibre content or presence of antinutrients can affect digestion. Therefore, the decreased performance at 1.5% might not be a toxic effect but rather a diminishing-return effect as compared to the 0.5% to 1.0% range.

There are some conflicting published results, emphasizing again the influence of dosage and experimental conditions. For instance, Alzawqari et al. (2016) concluded that body weight gain, feed intake and FCA were not significantly different among the groups but dried sweet orange peel supplementation significantly improved the antioxidant status and some serum variables during finisher phase. Differences between that study and the present one might be due to differences in the age of the birds, length of supplementation, preparation of the citrus, the chemical makeup, the formulation of the diets, environmental conditions, and inclusion rate. Unfortunately, there is not a standard that can be adhered to by all studies, so the differences between the studies do not necessarily reflect a contradiction, but rather show the chemical diversity of citrus products as a feed ingredient.

The results obtained from the effect on the serum lipid profile of the different dietary regimes with orange peel powder are presented in Figs. 1–5. In general, the levels of triglycerides (TG), total cholesterol (TC), HDL-C, VLDL-C and LDL-C were not significantly different between the different dietary treatments ($P > 0.05$). However, there were some number differences between treatments. Serum triglyceride concentration was 60.10 ± 3.21 in the Control-D1 group and rose numerically to 67.17 ± 4.71 in Group-D2 and 73.23 ± 6.68 in Group-D3. The highest mean TG level is thus observed in Group-D3 and the lowest numerical value was observed in the supplemented group, Group-D4 (59.54 ± 0.89), which is very close to the control. These differences, however, were not statistically significant ($P = 0.146$).

The same was observed for total cholesterol. The mean TC concentration was 73.99 ± 5.73 in Control-D1, 78.60 ± 2.71 in Group-D2, 98.11 ± 10.73 in Group-D3, and 74.49 ± 10.57 in Group-D4. However there was relatively high within group variation in Group-D3 and the overall treatment effect was not significant ($P = 0.176$), though the numerical mean was higher. For HDL-C, only minor differences appeared among treatments: the averages were 32.00 ± 1.41 in Control-D1, 31.50 ± 4.33 in Group-D2, 29.00 ± 4.14 in Group-D3, and 33.50 ± 2.40 in Group-D4. The numerical HDL-C level in Group-D4 was the highest, and in Group-D3, the lowest; however, there was no significant difference among groups ($P = 0.810$).

VLDL-C followed a pattern comparable to that of triglycerides, increasing from 12.02 ± 0.64 in Control-D1 to 13.43 ± 0.94 in Group-D2 and 14.65 ± 1.34 in Group-D3, then declining to 11.91 ± 0.18 in Group-D4. The treatment effect was not significant ($P = 0.146$). Similarly, serum LDL-C was 29.97 ± 4.32 in Control-D1, 33.67 ± 4.54 in Group-D2, 54.46 ± 12.06 in Group-D3, and 29.09 ± 9.10 in Group-D4. The concentration of LDL-C was highest in group-D3, although the standard error for the group was relatively large, indicating considerable variation within the replicates. As a result, the differences among dietary treatments were not significant ($P = 0.149$).

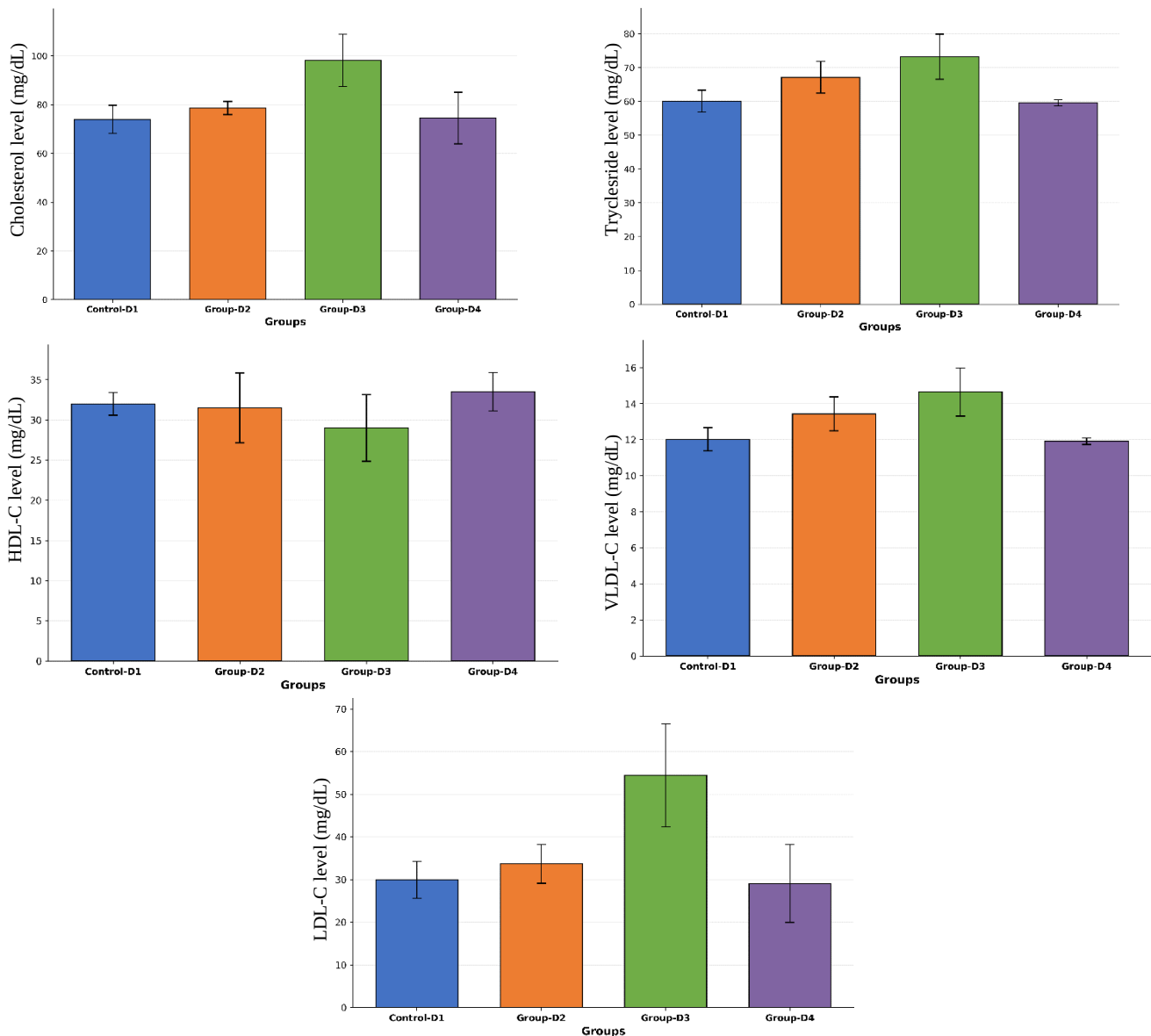


Figure 1. Effect of Dietary Peel Powder Supplementation on Serum Liquid Profile are Presented in Figures 1-5. Overall, Dietary Treatment Did Not Significantly Affect Triglycerides (TG), Total Cholesterol (TC), HDL-C, VLDL-C, or LDL-C among the Experimental Groups ($P>0,05$)

The results of this study show that adding orange peel powder to broiler diets did not significantly affect TG, TC, HDL-C, VLDL-C and LDL-C. Group-D3 did show numerically higher TG, TC, VLDL-C and LDL-C levels than the others but the heterogeneity of individual replicates, especially in TC and LDL-C, prevented these differences from being statistically significant (Figure-1). These numerical increases in the 1.0% orange peel group should not be taken as evidence of an adverse effect. Evidence of a dose-dependent response to orange peel powder is also lacking. Group-D2 showed only slight numerical increases in most lipid fractions and Group-D3 had the highest TG, TC, VLDL-C and LDL-C values. However, by increasing the inclusion level of the group to 1.5% for Group-D4 the values were close to those of the control group. This nonlinear pattern suggests that the effect of orange peel on lipid

metabolism is both in terms of its inclusion level and the composition of fiber fraction and biologically active compounds. Orange peel contains a range of bioactive ingredients, such as hesperidin and naringin, phenolic compounds, pectin, carotenoids and essential oils. Citrus flavonoids are known to affect lipid metabolism in hepatic lipid synthesis, fatty acid oxidation, antioxidant activity and lipid transport. Poultry nutrition has been extensively reviewed and found that flavonoids in food may affect fat deposition and serum lipid metabolism, but the response depends on the type of compound, the dose, the birds' physiological status and the food. There are several previous studies on the lipid-lowering effect of orange peel supplementation and Alzawqari et al. (2016) found that the dried sweet orange peel was beneficial for broilers in terms of serum triglycerides, LDL and VLDL and also for antioxidant status in the same way Akbarian et al. (2016) did. In a 2015 study, it was observed that the dietary orange peel extract at 400 mg/kg under heat stress reduced the serum LDL and cholesterol levels significantly. Abbasi et al. (2015) measured dried sweet orange residues at 0.5, 1.0, 1.5 and 2.0% of the diet, which were the same as those used in the present study. The highest inclusion level significantly decreased serum triglycerides as compared with the control and lower levels.

Not all published studies observe a uniform lipid-lowering response, though. Abd El Latif et al. (2023) found that orange peel meal had a significant effect on serum cholesterol, triglycerides, HDL and VLDL in broilers but the direction of the effect varied with the inclusion level and LDL did not change significantly. In some treatments, for instance, cholesterol and HDL increased while TG or VLDL decreased. This is consistent with the above findings and shows that citrus by-products can change serum lipid metabolism in a complex (not linear) way, not only linear. One explanation for the favorable lipid responses seen in other studies is that citrus flavonoids are involved. Hesperidin, naringin and related compounds have been shown to affect lipid synthesis, fatty acid oxidation, and cholesterol metabolism in poultry and in other experimental models. Tan et al. (2022) have found that dietary flavonoids can affect lipid metabolism in poultry and may reduce cholesterol and triglyceride levels but these effects depend on the dose and the flavonoid used. Our data show that the levels of orange peel powder used here were insufficient to produce a consistent hypolipidemic effect in serum. Another reason may be that whole peel contains bioactive flavonoids and a large fiber fraction, so the effective amount of individual compounds such as hesperidin or naringin may be much lower than in studies done with concentrated extracts. This is especially true when comparing whole orange peel powder with purified extracts. The very large numerical increase in LDL-C and TC in Group-D3 should also be taken into account in the future studies because of the large standard errors. Such high variability in within-group study is problematic in detecting reliable treatment effect and indicates that the increase was not consistent in all birds. It would be interesting to see if this pattern is a real biological response or a normal individual variation. Group-D4 showed TG, TC, VLDL-C and LDL-C close to those of control and numerically highest HDL-C concentration. Therefore, it may be possible that increasing the orange peel level to 1.5% did not affect lipid metabolism. Since the differences were not statistically significant, though, this is just a numerical trend and not a good effect. Overall, the lipid profile results indicated that we can include orange peel powder in broiler diets at 0.5-1.5% without any significant changes in the circulating lipid parameters. The absence of significant increases in TC, TG, LDL-C or VLDL-C also indicates that these inclusion levels did not change lipid metabolism in a consistent manner. In future studies with larger sample sizes, liver

lipid content, antioxidant status and hepatic enzyme activity measurements in addition to quantification of orange peel flavonoids could help to understand the physiological conditions driving these findings.

Conclusion

The results of the present performance indicate that 0.5% of orange peel powder was especially effective in increasing final body weight, and 1.0% was most effective when it comes to the overall feed conversion. A higher inclusion figure (1.5%) did not result in further improvements indicating a nonlinear response. Based on these performance parameters alone, the findings suggest that moderate inclusion levels of orange peel powder are appropriate, however more digestibility, intestinal morphology, antioxidant status and gut microbiota parameters would be required to confirm the physiological mechanisms responsible for the observed effects.

REFERENCES

- Abbasi, H., Seidavi, A., Liu, W., & Asadpour, L. (2015). Investigation on the effect of different levels of dried sweet orange (*Citrus sinensis*) pulp on performance, carcass characteristics and physiological and biochemical parameters in broiler chicken. *Saudi Journal of Biological Sciences*, 22(2), 139–146. <https://doi.org/10.1016/j.sjbs.2014.09.006>
- Abd El Latif, M. A., Abdel-Wareth, A. A. A., Daley, M., & Lohakare, J. (2023). Effect of dietary orange peel meal and multi-enzymes on productive, physiological and nutritional responses of broiler chickens. *Animals*, 13(15), 2473. <https://doi.org/10.3390/ani13152473>
- Abdelli, N., Solà-Oriol, D., & Pérez, J. F. (2021). Phytogetic feed additives in poultry: Achievements, prospective and challenges. *Animals*, 11(12), 3471. DOI: 10.3390/ani11123471.
- Akbarian, A., Golian, A., Kermanshahi, H., De Smet, S., & Michiels, J. (2015). Antioxidant enzyme activities, plasma hormone levels and serum metabolites of finishing broiler chickens reared under high ambient temperature and fed lemon and orange peel extracts and *Curcuma xanthorrhiza* essential oil. *Journal of Animal Physiology and Animal Nutrition*, 99(1), 150–162. <https://doi.org/10.1111/jpn.12188>
- Alzawqari, M. H., Al-Baddany, A. A., Al-Baadani, H. H., Alhidary, I. A., Khan, R. U., Aqil, G. M., & Abdurab, A. (2016). Effect of feeding dried sweet orange (*Citrus sinensis*) peel and lemon grass (*Cymbopogon citratus*) leaves on growth performance, carcass traits, serum metabolites and antioxidant status in broiler during the finisher phase. *Environmental Science and Pollution Research*, 23(17), 17077–17082. <https://doi.org/10.1007/s11356-016-6879-7>
- Alzawqari, M. H., Moghaddam, H. N., Kermanshahi, H., & Raji, A. R. (2013). Effect of feeding citrus peel extracts on growth performance, serum components, and intestinal morphology of broilers exposed to high ambient temperature during the finisher phase. *Livestock Science*, 157(2–3), 490–497. <https://doi.org/10.1016/j.livsci.2013.08.010>
- AOAC International. *Official Methods of Analysis of AOAC International*.
- Aviagen. *Ross 308 Broiler: Nutrition Specifications*. Aviagen Group.
- Cisse, S., Bahut, M., Marais, C., Zemb, O., Chicoteau, P., Benarbia, M. E. A., & Guilet, D. (2023). Fine characterization and microbiota assessment as keys to

- understanding the positive effect of standardized natural citrus extract on broiler chickens. *Journal of Animal Science*, 101, skad069. DOI: 10.1093/jas/skad069.
- Liu, R., Ding, X., Dang, M., Wang, J., & Zhu, W. (2024). Effects of hesperidin, thymol, rosmarinic acid and their combined effect on growth performance, intestinal barrier function and cecal microbiota in broilers. *Poultry Science*, 103(12), 104247. DOI: 10.1016/j.psj.2024.104247.
- Mahfuz, S., et al. (2022). Dietary the effects of graded dietary levels of orange peel powder on productive performance and selected physiological traits in broiler chickens.
- Mulvihill, E. E., Burke, A. C., & Huff, M. W. (2016). Citrus flavonoids as regulators of lipoprotein metabolism and atherosclerosis. *Annual Review of Nutrition*, 36, 275–299. <https://doi.org/10.1146/annurev-nutr-071715-050718>
- Ogunlade, J. T., Chukwudi, P., Gabriel, G. O., Akingbade, T. F., Adaramodu, T. T., Adesokan, E. D., Bawa, N. T., Owolabi, S. J., Adebayo, M. K., Olatundun, B. E., & Ogwuegbu, M. C. (2025). Incremental dietary supplementation with sweet orange (*Citrus sinensis*) peel meal on the growth performance, carcass traits, haematology, serum biochemistry, electrolyte balance, antioxidant status, and humoral immunity in broiler chickens. *Discover Life*, 55, 37. <https://doi.org/10.1007/s11084-025-09716-2>
- Polycarpo, G. do V., de Lima, G. A., Ávida, T. de S., Rosas, F. S., Cruz-Polycarpo, V. C., Biller, J. D., Barbosa, B. F. da S., & Burbarelli, M. F. de C. (2022). Can phytogenic additives improve the performance of broilers and replace growth-promoting antibiotics? A meta-analytic approach. *Canadian Journal of Animal Science*, 102(2), 289–300. DOI: 10.1139/cjas-2021-0058.
- Sugiharto, S. (2023). The effect of using fruit peel on broiler growth and health. *Veterinary World*, 16(5), 987–1000. DOI: 10.14202/vetworld.2023.987-1000.
- Tan, Z., Halter, B., Liu, D., Gilbert, E. R., & Cline, M. A. (2022). Dietary flavonoids as modulators of lipid metabolism in poultry. *Frontiers in Physiology*, 13, 863860. <https://doi.org/10.3389/fphys.2022.863860>
- Vlaicu, P. A., Untea, A. E., Panaite, T. D., & Turcu, R. P. (2020). Effect of dietary orange and grapefruit peel on growth performance, health status, meat quality and intestinal microflora of broiler chickens. *Italian Journal of Animal Science*, 19(1), 1394–1405. DOI: 10.1080/1828051X.2020.1845576.