Journal of Human Environment and Health Promotion
Print ISSN: 2476-5481 Online ISSN: 2476-549X

Effects of *Amaranthus hybridus chlorostachys* Feeding on Productivity in Broiler Chickens: An Animal Model Study

Mahmoud Saei [a], **Alireza Seidavi** [a *], **Mehrdad Bouyeh** [a]

a. Department of Animal Science, Faculty of Agriculture, Ra.C., Islamic Azad University, Rasht, Iran.

***Corresponding author:** Department of Animal Science, Faculty of Agriculture, Ra.C., Islamic Azad University, Rasht, Iran. Postal Code: 41335-3516. E-mail: alireza.seidavi@iau.ac.ir

ARTICLE INFO

Article type:
Original article

Article history:
Received: 19 August 2025
Revised: 9 September 2025
Accepted: 25 September 2025
Available online: 6 October 2025

https://doi.org/10.61882/jhehp.719

Keywords:

Amaranth
Atherogenic index
Broiler chickens
Enrichment
Feed conversion ratio
Total antioxidant capacity

ABSTRACT

Background: This study investigated the effects of amaranth feeding on performance, carcass characteristics, biochemical parameters, blood antioxidant status, immunity, fatty acid profile, and sensory traits of broiler meat.
Methods: After analyzing the nutrient composition of amaranth, experiments were conducted using 150 Ross 308 broilers in a completely randomized design with 3 treatments (0, 1, and 2% raw amaranth) and five replications of 10 birds each.
Results: Results showed that broilers fed 1% or 2% amaranth exhibited an improved feed conversion ratio and increased body weight ($p < 0.05$). Amaranth feeding also decreased ($p < 0.01$) blood glucose, cholesterol, low-density lipoprotein, atherogenic index, alanine aminotransferase, aspartate aminotransferase, and malondialdehyde. However, it significantly increased high-density lipoprotein, uric acid, albumin, total protein, phosphorus, iron, calcium, and total antioxidant capacity ($p < 0.01$).
Conclusion: Supplementation with low levels of amaranth can be used as a dietary enrichment for broiler chickens without negative effects on overall production performance or carcass traits. Improvements were observed in health-related indicators, including blood lipid and antioxidant profiles, as well as the sensory quality of broiler meat. Since broiler meat is widely consumed, this research suggests a potential benefit for human health.

1. Introduction

The use of non-traditional edible plants for dietary supplementation has the potential to be a low-risk and acceptable change in today's poultry industry (Hosseintabar-Ghasemabad et al., 2024). Therefore, nutritionists are constantly trying to enhance the success of the poultry industry by investigating alternative plant-based feed sources and incorporating them into diet formulations (Phillips et al., 2023).

Amaranth is considered a valuable food plant that is similar to and close to the cereal family in terms of energy values (i.e., it is a pseudocereal), and, in parallel, its other nutritional, micronutrient, and bioactive compounds have a significant advantage compared to cereals (Patro & Chowdhury, 2025). According to the Food and Agriculture Organization of the United Nations (FAO, 2023), the production of amaranth grain was nearly 2 million tons in 2023. With the increasing demand for organic products and gluten-free cereals, its production is estimated to reach $2.5 billion by 2030 with an annual growth rate of 8-10% (Azri et al., 2025).

Because poultry require a wide range of nutrients to meet their dietary needs, supplementing rations with alternative feedstuffs such as amaranth grain represents a promising dietary modification. A review of the scientific literature has indicated the effectiveness of amaranth in broiler chickens, laying hens, Japanese quail, breeder hens, and geese; however, the optimal inclusion level for poultry diets needs further investigation to maximize flock performance and

How to cite: Saei, M., Seidavi, A., & Bouyeh, M. (2025). Effects of *Amaranthus hybridus chlorostachys* Feeding on Productivity in Broiler Chickens: An Animal Model Study. *J Hum Environ Health Promot*, *11*(4), 225-237.

health while minimizing production costs (Janmohammadi et al., 2022).

Botanical references (Flora Iranica, 2025; Mozaffarian, 1996) reveal that the studied amaranth (*Amaranthus hybridus chlorostachys*) is an annual herbaceous plant, 30 to 100 cm high (sometimes more), pubescent on the upper part, with oblong-ovate and lanceolate leaves, with a blunt, beaked and prickly apex, pubescent on the lower surface, pubescent along the veins, axial pseudo spikes, and in general, a leafless inflorescence-a branched panicle, olive green to pale green, and seeds about 1 mm in diameter and dark in color. *A. hybridus* is considered a cosmopolitan species that has good germination and is more accessible in many parts of the world (Nwadinigwe et al., 2019). Compared to soybeans, amaranth is a drought-resistant crop that has a higher tolerance threshold. In dry conditions, it can temporarily wilt and recover immediately after rainfall with favorable efficiency.

The high content of lysine, arginine, and methionine in amaranth grain can be very important in poultry nutrition; however, leucine, valine, and threonine are limiting amino acids in amaranth grain, which can largely be eliminated in poultry nutrition by feeding corn and soybean meal (Bejosano & Corke, 1998). A review of the scientific literature indicates the functional potential of using amaranth in feeding broiler chickens as a beneficial source of energy and protein in poultry diets. In addition to meeting the nutritional needs of poultry and eliminating possible deficiencies, it can also play a role as a fortifier in improving health and modulating immunity due to its valuable bioactive compounds (Seidavi et al., 2023).

Feeding extruded amaranth to broilers at levels of 0, 10, 20, 30, 40, and 50% (Tillman & Waldroup, 1988) could have a positive effect up to 40% of the diet without any negative effect on broiler body weight, performance, carcass characteristics, and feed conversion ratio (FCR). In two separate experiments the nutritional value of raw and autoclaved *A. hypochondriacus* in broiler chickens′ diets at levels of 0, 20, 40 and 60% was examined (Ravindran et al., 1996); from days 6 to 17 of age, increasing levels of raw amaranth led to a decrease in feed intake and body weight and impaired FCR, but feeding autoclaved amaranth eliminated the negative growth-reducing effects such that levels of 20 and 40% had similar performance to the control group. Another investigation (Rouckova et al., 2004) observed performance similar to the control group when feeding processed amaranth grain up to 7% as a replacement for meat and bone meal, although blood biochemical parameters indicated a decrease in blood glucose and lipids. Supplementing broiler chickens' diets with *A. caudatus* grain at three levels (0, 5, and 10% showed that, with increasing levels of raw amaranth feeding, there was a noticeable decrease in performance, but parameters related to improving flock health, including antioxidant capacity and serum lipid peroxidation, improved (Longato et al., 2017).

While Amaranth (commonly, *Amaranthus* spp.) and its grains are valued worldwide for nutritional benefits, "amaranth" is also the name of a synthetic azo dye (E123) previously used as a food coloring. In Iran, synthetic amaranth colorant is not authorized for use in foods, with national standards highlighting concerns over potential toxicological effects. However, it is vital to differentiate between the unauthorized synthetic colorant "Amaranth" and the botanical *Amaranthus hybridus* used in this study, which pertains to the edible grain, widely recognized as a functional food ingredient in many cultures.

The findings of this research suggest an avenue to promote the health of humans who consume broiler meat. Based on the results of the current study, we can next evaluate the amaranth effects in human nutrition. The nutritional quality and safety of poultry meat are pivotal public health concerns, as chicken represents a primary protein source worldwide. Improvements in poultry feed composition can translate directly into healthier meat for consumers, promoting broader population health outcomes. Integrating functional ingredients such as *Amaranthus hybridus chlorostachys* into broiler diets not only affects animal growth and efficiency but also enhances the nutritional and biochemical profile of the resultant meat. Amaranth grains, rich in bioactive compounds like squalene, polyphenols, and unsaturated fatty acids, modulate lipid metabolism and antioxidant status in monogastric animals. These changes may be reflected in the meat consumed by humans, potentially contributing to reduced cardiovascular risk factors and supporting disease prevention, which is a public health priority.

The present study addresses the intersection between food animal nutrition and human health promotion by investigating whether the low-level inclusion of amaranth in poultry feed can yield carcass and meat products with improved nutritional profiles, without impairing broiler productivity. Thus, the findings have implications for food safety, nutrition, and the development of functional animal products, core areas within the topic of human health promotion. Due to positive reports about the effects of amaranth (*Amaranthus hybridus chlorostachys*) in some organisms, we examined its feeding effects on production performance, carcass characteristics, biochemical parameters, and blood antioxidant status, immune system, fatty acid profile, and sensory and taste traits of meat in broiler chickens as an animal model.

2. Materials and Methods

2.1 Preparation and analysis of test ingredients

The present study investigated the potential of grain amaranth (*Amaranthus hybridus chlorostachys*), obtained from Darvash Giah Khazar Medicinal Herbs Complex (Rasht, Guilan, Iran), as a feed ingredient for broilers. A sample of the grain amaranth underwent a series of nutritional and micronutrient compound analyses in laboratories approved by the Iranian Food and Drug Organization, including Viromed (Rasht, Guilan, Iran) and Tekno Azma (Iran, Tehran).

Based on the Iranian National Standard Method (ISIRI protocol), 18 parameters were analyzed, including protein (Method 10703-1), fat (Method 10700), crude fiber (Method

3105), ash (Method 11143), dry matter (Method 8438), neutral-detergent fiber (NDF; Method 8917), acid detergent fiber (ADF; Method 8917), total sugar (Method 8986-2), starch (Method In House), total phenolic compounds (Method 8986-1), calcium (Method 10701-1), phosphorus (Method 513), magnesium (Method In House), iron (Method In House), phosphorus (Method In House), vitamin A (Method ISIRI 7432), vitamin D3 (Method ISIRI 13579), vitamin K3 or menadione (Method In House) and amino acid profile (Method ISO 13903).

In addition, the fatty acid profile (Method 13126-1-2) and squalene content (Method 9670) were determined according to the Iranian National Standard Method (AOAC protocol). Nitrogen-free extract (NFE) and Nonfibrous carbohydrates (NFC) were also calculated and are reported in Table 1, along with all laboratory values.

Table 1. Nutrient and micronutrient composition of amaranth grain (*Amaranthus hybridus chlorostachys*) fed to broilers at 1% or 2% of the diet

Items	Value	Items	Value (g/100g)	Items	Value (%)
Crude Protein (%)	16.6	Proline	0.3	C12:0	0.11
Crude fat (%)	7.06	Taurine	0.2	C14:0	0.29
Crude fiber (%)	12.87	Tryptophan	<0.1	C16:0	16.35
Ash (%)	6.50	Lysine	1.15	C16:1	0.39
Dry matter (%)	91.9	Leucine	0.11	C17:0	0.14
Starch (%)	11.36	Isoleucine	0.49	C17:1	0.78
NDF (%)	37.40	Phenylalanine	0.12	C18:0	4.38
ADF (%)	29.50	Valine	<0.04	C18:0	25.00
Total phenolic compounds (%)	9.53	Methionine	<0.06	C18:1	50.14
Calcium (%)	0.59	Tyrosine	<0.26	C18:2	0.53
Phosphorus (%)	0.55	Alanine	<0.04	C18:3	0.92
Magnesium (%)	0.38	Arginine	<0.07	C20:0	0.30
Iron (mg/kg)	78	Threonine	<0.04	C20:1	0.31
Vitamin A (mg/kg)	0.21	Glycine	<0.03	C22:0	0.18
Vitamin D_3 (mg/kg)	0.30	Histidine	<0.06	C24:0	0.05
Vitamin K_3 (mg/kg)	0.30	Serine	<0.05	C24:1	0.07
NFE (%)	56.97	Glutamic acid	<0.05	C20:2	0
NFC (%)	32.44	Aspartic acid	<0.05	Squalene (ppm)	2550.39
NFE (%) = 100 – (Crude protein + Crude fat + Crude fiber + Ash)					
NFC (%) = 100 – (Crude protein + Crude fat + NDF + Ash)					

NFE (%) = 100 – (Crude protein + Crude fat + Crude fiber + Ash)
NFC (%) = 100 – (Crude protein + Crude fat + NDF + Ash)

2.2 Broiler Management and Breeding

The present study was conducted using 150 Ross 308 broiler chickens with a similar initial average weight (41±1 g). A completely randomized design was applied with three dietary treatments (0, 1, and 2% raw amaranth powder). Each treatment was replicated 5 times with 10 birds in land cages having dimensions of 1.2m×1.5m×2.0m.

The research was conducted at the Research and Development Farm owned by Sepid Makian Company (Someh-Sara, Guilan, Iran), and encompassed the three phases of broiler production: starter (1-11 d), grower (12-21 d), and finisher (22-42 d).

All stages of broiler breeding and sampling were carried out following the welfare guidelines in accordance with the Ethics Committee of the Islamic Azad University, Rasht Branch (ID: IR.IAU.RASHT.REC.1402.021). Husbandry conditions (temperature, lighting, creating conditions for free access to drinking water and feed, and the vaccination schedule) were maintained according to the breeder's management guide (Manual, 2012).

The experimental diets were based on corn and soybean meal and formulated using Amino Feed 5.0 software (Evonik Digital GmbH, Essen, Germany; https://animal-nutrition.evonik.com/en) as shown in Table 2. The metabolic energy values (AME_n) of the studied amaranth were also considered to be 3255 kcal/kg for diet writing, based on the report of Janmohammadi et al. (2022), and the reason for this was the commonness of the food source in the present study.

2.3 Parameters measured

2.3.1 Performance parameters

Broiler performance was evaluated during three periods (starter, grower, and finisher) by measuring feed intake, body weight, and calculating FCR. In addition, on the last day of rearing (42 d), after four hours of starvation, two birds were randomly selected from each pen (each replication) and slaughtered. The average pre-slaughter live weight of the selected birds was considered close to the average weight of the chicks in that experimental unit. After carcass separation, the measured components included the following weights, taken with a digital scale having an accuracy of 0.001 g: full carcass, defeathered body, eviscerated carcass, breast, thigh, gizzard, crop, liver, heart, pancreas, spleen, bursa of Fabricius, and abdominal fat.

2.3.2 Blood parameters

On the last day of rearing, three birds were randomly selected from each replicate, and blood samples were collected from the wing vein using 5 cc sterile syringes. All samples were centrifuged (3000 rpm) in cool conditions to obtain serum. For analysis of the desired parameters, all samples were analyzed in a commercial laboratory (Rasht-Guilan, Iran) using commercially available kits.

Blood biochemical parameters were measured based on specialized laboratory protocols with the Colorimetric

method for glucose, triglyceride (TG), total cholesterol (TC), high density lipoprotein (HDL), low density lipoprotein (LDL), total protein (TP), albumin (Alb), uric acid (UA), calcium (Ca), iron (Fe), total antioxidant capacity (TAC), and malondialdehyde (MDA). In addition, phosphorus (P) was measured by the Photometric method, and aspartate aminotransferase (AST) and alanine aminotransferase (ALT) were measured by the Enzymatic method. The atherogenic index (AI) was also calculated as an important indicator for health, based on the ratio of LDL to HDL.

Table 2. Components and nutrient compositions of the three experimental diets (T1, T2, and T3) 1 fed to broilers during the starter, grower, and finisher periods

Items	Starter (1-11 d)			Grower (12-21 d)			Finisher (22-42 d)		
Ingredients (%)	**T_1: 0**	**T_2: 1**	**T_3: 2**	**T_1: 0**	**T_2: 1**	**T_3: 2**	**T_1: 0**	**T_2: 1**	**T_3: 2**
Corn	53.54	52.66	52.29	61.78	61.20	60.63	67.59	67.01	66.43
Amaranth	0.00	1.00	2.00	0.00	1.00	2.00	0.00	1.00	2.00
Soybean meal 44%	40.95	40.78	40.46	33.23	32.98	32.73	27.35	27.09	26.84
Vegetable oil	1.53	1.53	1.21	1.39	1.21	1.04	1.91	1.73	1.56
Methionine	0.35	0.35	0.35	0.28	0.28	0.28	0.22	0.22	0.22
Lysine hydrochloride	0.21	0.21	0.22	0.21	0.21	0.22	0.20	0.21	0.21
Threonine	0.11	0.11	0.11	0.09	0.09	0.09	0.07	0.07	0.08
Valine	0.04	0.04	0.04	0.02	0.02	0.02	0.01	0.01	0.01
Choline chloride	0.01	0.05	0.05	0.07	0.07	0.07	0.07	0.07	0.07
Monocalcium phosphate	1.11	1.11	1.11	0.90	0.90	0.90	0.69	0.69	0.69
Calcium carbonate	1.19	1.19	1.19	1.06	1.06	1.06	0.92	0.92	0.92
Sodium bicarbonate	0.24	0.26	0.26	0.27	0.27	0.27	0.27	0.27	0.28
Sodium chloride	0.21	0.20	0.19	0.19	0.19	0.19	0.19	0.19	0.19
Vit and Min Premix[2]	0.50	0.50	0.50	0.50	0.50	0.50	0.50	0.50	0.50
Phytase	0.005	0.005	0.005	0.005	0.005	0.005	0.005	0.005	0.005
ME (kcal/kg)[3]	2853	2853	2853	2937	2937	2937	3040	3040	3040
Crude protein	22.91	22.94	22.98	19.98	20.10	20.04	17.70	17.74	17.77
Lysine	1.26	1.26	1.26	1.09	1.09	1.09	0.96	0.96	0.96
Met + Cys[4]	0.94	0.94	0.94	0.81	0.81	0.81	0.71	0.71	0.71
Threonine	0.83	0.83	0.83	0.72	0.72	0.72	0.63	0.63	0.63
Tryptophan	0.25	0.25	0.25	0.21	0.21	0.21	0.18	0.18	0.18
Arginine	1.40	1.40	1.40	1.20	1.19	1.19	1.04	1.03	1.03
Isoleucine	0.86	0.86	0.86	0.74	0.74	0.74	0.65	0.65	0.65
Valine	0.97	0.97	0.97	0.84	0.84	0.84	0.74	0.74	0.74
Calcium	0.95	0.95	0.95	0.84	0.84	0.84	0.73	0.73	0.73
Av. Phosphorus[5]	0.48	0.48	0.48	0.42	0.42	0.42	0.37	0.37	0.37
Sodium	0.16	0.16	0.16	0.16	0.16	0.16	0.16	0.16	0.16
Potassium	1.01	1.01	1.00	0.88	0.88	0.87	0.78	0.77	0.77
Chlorine	0.21	0.21	0.21	0.21	0.21	0.21	0.21	0.21	0.21
DCAB (mEq/kg)	269	269	268	235	235	234	209	208	208
Choline (g/kg)	1.47	1.69	1.69	1.59	1.59	1.59	1.48	1.48	1.48
Ether extract	4.19	4.18	3.87	4.24	4.06	3.88	4.87	4.69	4.51
Linoleic acid	1.99	1.98	1.82	2.02	1.93	1.84	2.34	2.25	2.16
NDF	9.65	9.66	9.72	9.70	9.74	9.78	9.70	9.74	9.78
ADF	4.19	4.19	4.19	3.97	3.97	3.97	3.79	3.79	3.79
Crude fiber	3.10	3.10	3.10	2.94	2.94	2.94	2.81	2.81	2.81
Ash	6.17	6.17	6.17	5.44	5.44	5.43	4.77	4.77	4.77
Starch	34.20	34.24	34.61	39.38	39.62	39.85	43.03	43.27	43.50

[1]T_1: control group: 0% amaranth, T_2: 1% amaranth, T_3: 2% amaranth; [2]The values of vitamins and minerals per kg of the assay diet: Vitamin A, 9000 IU; vitamin D_3, 3000 IU; vitamin E, 18 IU; vitamin K_3, 3 mg; vitamin B_1, 1.8 mg; vitamin B_2, 6 mg; vitamin B_6, 3 mg; vitamin B_{12}, 0.012 mg; vitamin B_3, 30 mg; vitamin B_9, 1 mg; vitamin H_3, 0.24mg; vitamin B_5, 10 mg; 500 mg; Choline,100 mg; Mn, 100 mg; Zinc, 80 mg; Iron, 10 mg; Cu, 1 mg; [3]ME: Metabolizable Energy; [4]Methionine + Cysteine; [5]Av. phosphorus: Available Phosphorus

2.3.3 Immunological parameters

In order to evaluate immune system parameters, two birds from each replicate were selected for injection and sampling on days 28 and 36 of age. 0.2 cc of a 5% dilution of sheep red blood cells (SRBC) was injected into the pectoral muscle. Seven days after SRBC injection, on days 35 and 42, blood samples were collected from the wing vein of the same birds (Amirdahri et al., 2023) using 3-cc sterile syringes. The levels of antibodies against SRBC were measured using the hemagglutination method to assess Newcastle disease (NDV) and influenza (AIV) titers. Organs related to the immune system (spleen, bursa of Fabricius) were weighed, and humoral immunity was assessed. After injection and sampling, all immunological laboratory assays were conducted in the laboratory (Rasht-Guilan, Iran).

2.3.4 Meat fatty acid parameters

After slaughtering birds at the end of the finisher period, one whole chicken breast was separated from one bird from each treatment and transferred to the laboratory to evaluate the fatty acid profile of the meat.

Breast tissue samples were prepared as a homogeneous mixture with 100 mL of methanol: chloroform (2:1) for four hours, and then the samples were filtered and mixed with 25 mL of saturated sodium chloride solution in a decanter funnel. The chloroform phase (containing the fat) was filtered using filter paper impregnated with anhydrous potassium sulfate. The filtered samples were dried by a rotary evaporator under vacuum so that only fat remained. Ten mg of extracted fat were mixed with two mL of potassium hydroxide, two mL of normal methanol, and seven

ml of n-hexane, and the samples were subsequently centrifuged for 10 min. Samples were left in a stationary state for five minutes until the supernatant phase was separated, and then approximately one µl of the supernatant was injected into a gas chromatography device to evaluate the fatty acid profile, and the amount of the above fatty acids was expressed as a percentage.

A series of health-promoting indices related to fatty acids, including the ratio of omega-6 to omega-3 fatty acids, the atherogenic index (AI), the thrombogenic index (TI), the hypocholesterolemic index (HI), and the hypocholesterolemic to hypercholesterolemic ratio, were calculated using previously published formulas (Attia et al., 2022):

Formula (1): AI = (4 × C14:0) + C16:0 / (ΣMUFA + ΣPUFA – ω-6 + ΣPUFA – ω-3)

Formula (2): TI = (C14:0 + C16:0 + C18:0) / 0.5 × Σ MUFA + 0.5 × Σ (ω-6) + 3 × Σ (ω-3) + Σ (ω-3) / Σ (ω-6)

Formula (3): HI = (C18:1 + C18:2 + C18:3 + C20:3 + C20:4 + C20:5 + C22:4 + C22:6) / (C14:0 + C16)

Formula (4): Hypocholesterolemic / Hypercholesterolemic index = [(C18:1 ω-9 + C18:1 ω-7 + C18:2 ω-6 + C18:3 ω-6 + C18:3 ω-3 + C20:3 ω-6 + C20:4 ω-6 + C20:5 ω-3 + C22:4 ω-6 + C22:5 ω-3 + C22:6 ω-3) / (C14:0 + C16:0)]

2.3.5 Sensory and taste parameters of breast meat

To evaluate the sensory and taste attributes of the broiler breast meat, at the end of the finisher period, a whole breast from each replicate was cooked without spices at 180 °C for 45 min. The cooked samples were evaluated by six panelists (food testers). Evaluators completed a questionnaire by assigning a score from 1 to 10 for aroma, taste, odor, crispness, color, and overall desirability. In order to prevent carryover flavor interference during the evaluation process, panelists rinsed their mouths with lukewarm water before and after tasting each sample of breast meat.

2.4 Statistical analysis

All data obtained in the present study were collected systematically and entered into an Excel spreadsheet (Microsoft, Redmond, Washington, USA) for subsequent analysis using statistical software (SAS 9.3 Institute, Cary, North Carolina, USA). Comparisons of treatment means were made using Duncan's multiple range test. In addition, quadratic, linear, and orthogonal equations were used to examine linear and nonlinear equations. The Excel "Solver" extension was used to find the turning point of quadratic equations. Statistical significance was considered when the P-value was less than 0.05.

3. Results and Discussion

Results of broiler performance are shown in Table 3. Growth performance from 1 to 11 days of age indicated that broilers in the control group had heavier body weights and better feed conversion ($p < 0.05$) than broilers consuming 1% or 2% grain amaranth, which were not different from one another. When contrasting the control group with the pooled amaranth groups, FCR and body weight were also different ($p < 0.05$). There was a significant linear effect for body weight (y = -14.102x + 449.77 with a coefficient of determination of 0.75) and FCR (y = 0.0125x + 1.0754 with a coefficient of determination of 0.47) concerning the percentage of amaranth used in the diet ($p < 0.05$). Letter y is the target trait, and letter x is the amaranth level.

There was no difference in the performance of broilers in the control group and the groups fed with amaranth for growth performance at 12-21 days, 22-42 days, and 1-42 days for feed intake, body weight, and FCR. In addition, there were no effects of contrast, linear, and quadratic equations between the treatments ($p > 0.05$).

Table 3. Results of performance traits of broiler chickens in experimental groups

Treatments	Feed Intake (g)	Body Weight (g)	FCR
Starter period performance (1-11 d)			
T_1 (control)	485.20	454.40[a]	1.068[b]
T_2	470.40	426.40[b]	1.103[a]
T_3	465.74	426.20[b]	1.093[a]
SEM	8.249	6.994	0.009
p-value	0.259	0.022	0.025
Control Vs. Amaranth	0.116	0.007	0.009
Linear	0.121	0.015	0.042
Quadratic	0.625	0.131	0.044
Grower period performance (12-21 d)			
T_1	586.60	415.20	1.41
T_2	589.00	412.60	1.43
T_3	584.07	419.53	1.39
SEM	9.786	10.400	0.022
p-value	0.939	0.894	0.501
Control Vs. Amaranth	0.996	0.947	0.914
Linear	0.858	0.773	0.499
Quadratic	0.765	0.715	0.342
Finisher period performance (22-42 d)			
T_1	2763.4	1509.8	1.833
T_2	2884.4	1599.4	1.807
T_3	2846.1	1571.5	1.811
SEM	41.230	35.962	0.029
p-value	0.148	0.237	0.804
Control Vs. Amaranth	0.067	0.112	0.528
Linear	0.181	0.248	0.633
Quadratic	0.141	0.207	0.659
Entire period performance (1-42 d)			
T_1	3835.2	2379.4	1.613
T_2	3943.8	2438.4	1.618
T_3	3895.9	2417.3	1.612
SEM	47.061	34.303	0.016
p-value	0.299	0.489	0.936
Control Vs. Amaranth	0.168	0.272	0.804
Linear	0.380	0.450	0.932
Quadratic	0.200	0.359	0.729

* Means with the same letter for each column are not significantly different.
* T_1: control group: 0% amaranth, T_2: 1% amaranth, T_3: 2% amaranth.

The weight of the carcass and carcass components is shown in Table 4. Treatment has no effect ($p > 0.05$) on any of the carcass traits.

Table 4. Weight (in grams) of various carcass and carcass components of broiler chickens fed 0, 1, or 2% grain amaranth

Treatment	Live body	Defeather body	Eviscerated carcass	Breast	Thigh	Abdominal fat	Gizzard	Heart	Crop	Liver
T_1 (control)	2378.0	2002.0	1708.0	604.0	472.0	31.54	34.88	10.72	6.72	49.36
T_2	2436.0	2088.0	1822.0	678.0	502.0	18.60	32.56	9.64	6.64	49.08
T_3	2392.0	2046.0	1798.0	632.0	480.0	24.48	37.70	11.00	7.18	46.64
SEM	36.533	37.434	32.823	20.769	18.779	3.551	1.875	0.745	0.447	1.879
p-value	0.522	0.303	0.070	0.075	0.523	0.071	0.194	0.421	0.664	0.547
Control Vs. Amaranth	0.437	0.182	0.026	0.068	0.425	0.040	0.915	0.669	0.735	0.527
Linear	0.791	0.422	0.076	0.359	0.768	0.185	0.309	0.795	0.481	0.326
Quadratic	0.277	0.188	0.112	0.036	0.280	0.050	0.130	0.206	0.582	0.647

* Means with the same letter for each column are not significantly different.
* T1: control: 0% amaranth, T2: 1% amaranth, T3: 2% amaranth.

However, the contrast effect of the control treatment compared to the simultaneous effect of the two amaranth groups at 1 and 2% levels was significant ($p < 0.05$) for the eviscerated carcass (g) and abdominal fat (g) weights. In addition, a significant quadratic equation was observed for breast weight (g) and abdominal fat weight (g) as follows. Letter y is the target trait, and letter x is the amaranth level.

Breast weight (g) = y = -60x2 + 134x + 604 min = 1.12

Abdominal fat weight (g) = y = 9.41x2 - 22.35x + 31.5 min = 1.19

The results of the blood parameter analysis depicted in Table 5 revealed that in all traits (except triglycerides), there was a difference ($p < 0.01$) between the tested treatments. Also, in all traits (except blood triglycerides), a difference ($p < 0.01$) was observed between the control group and the contrast of amaranth effects (0% compared to 1% and 2% amaranth). For all traits except blood triglycerides, linear equations were significant ($p < 0.01$). For blood cholesterol, a difference ($p < 0.01$) was observed between the control treatment and each of the amaranth treatments, which were higher than the control group. For HDL and TAC, a difference ($p < 0.01$) was observed between the control treatment and each amaranth treatment, with a decrease in the percentage of amaranth in the diet, and the amount of these two parameters had a decreasing trend. For LDL, AI, ALT, and MDA, a downward trend ($p < 0.01$) was observed with increasing amaranth levels, so that the control treatment showed higher values compared to the 1% and 2% amaranth treatments, as well as the 1% treatment being higher than the 2% treatment. For uric acid, total protein, phosphorus, Fe, and Ca, the highest values were observed in broilers fed 2% amaranth, and a difference was also observed between the control treatment and the 1% treatment ($p < 0.01$). For blood albumin, increasing the amaranth level had a positive ($p < 0.01$) effect; the blood albumin value in the 2% group had a significant difference with the control treatment and 1% amaranth ($p < 0.01$). A difference ($p < 0.01$) was observed between the 1% amaranth treatment and the control treatment. For ALT, a difference ($p < 0.01$) was observed between the 2% treatment and the other two treatments. For the AST parameter, no difference ($p > .0.05$) was observed between the 1% treatment and the other two treatments.

Table 5. Results of blood biochemical parameters of broiler chickens

Items	Treatments			SEM	*p-value*	Control Vs. Amaranth	Linear			Quadratic
	T_1	T_2	T_3				*p-value*	Equal	R^2	
Glucose (mg/dl)	175.80[a]	170.80[b]	168.40[b]	2.1087	0.0047	0.0102	0.0289	y = -3.7x + 175.37	0.96	0.6238
Cholesterol (mg/dl)	182.00[a]	158.00[b]	155.60[b]	1.0100	0.0001	0.0001	0.0001	y = -13.2x + 178.4	0.81	0.0001
HDL (mg/dl)	36.50[b]	38.70[a]	39.42[a]	0.4985	0.0036	0.0012	0.0014	y = 1.46x + 36.747	0.92	0.2488
LDL (mg/dl)	99.74[a]	90.64[b]	83.52[c]	0.6667	0.0001	0.0001	0.0001	y = -8.11x + 99.41	0.98	0.2487
Atherogenic Index	2.73[a]	2.34[b]	2.12[c]	0.0346	0.0001	0.0001	0.0001	y = -0.3064x + 2.70	0.97	0.0771
Triglycerides (mg/dl)	75.70	73.26	74.12	1.2664	0.4122	0.2194	0.3950	-	-	0.3083
Uric Acid (mg/dl)	5.61[b]	5.61[b]	5.76[a]	0.0165	0.0001	0.0047	0.0001	y = 0.071x + 5.5897	0.74	0.0036
Albumin (g/dl)	2.06[c]	2.14[b]	2.22[a]	0.0136	0.0001	0.0001	0.0001	y = 0.079x + 2.0637	0.99	0.9531
Total Protein (g/dl)	4.41[b]	4.43[b]	4.82[a]	0.0129	0.0001	0.0001	0.0001	y = 0.205x + 4.3483	0.78	0.0001
Phosphorus (mg/dl)	4.14[b]	4.24[b]	5.13[a]	0.0621	0.0001	0.0001	0.0001	y = 0.495x + 4.0077	0.82	0.0002
Fe (µg/dl)	130.20[b]	133.40[b]	144.20[a]	2.0928	0.0012	0.0057	0.0005	y = 7x + 128.93	0.91	0.1640
Ca (mg/dl)	10.72[b]	10.75[b]	10.94[a]	0.0313	0.0005	0.0083	0.0003	y = 0.11x + 10.695	0.82	0.0405
ALT (u/l)	15.36[a]	11.44[b]	9.96[c]	0.4354	0.0001	0.0001	0.0001	y = -2.7x + 14.953	0.93	0.0411
AST (u/l)	351.80[a]	347.00[a]	299.80[b]	3.3257	0.0001	0.0001	0.0001	y = -26x + 358.87	0.83	0.0002
MDA (nmol/mL	19.16[a]	18.50[b]	18.04[c]	0.1485	0.0007	0.0004	0.0002	y = -0.56x + 19.127	0.98	0.5926
TAC (nmol/ml)	675.80[b]	721.80[a]	724.60[a]	9.8020	0.0067	0.0019	0.0042	y = 24.4x + 683	0.79	0.0972

* Means with the same letter for each row are not significantly different.
* T1: control group: 0% amaranth, T2: 1% amaranth, T3: 2% amaranth.

Immune system-related traits in broiler chickens, shown in Table 6, indicated that treatment affected ($p < 0.05$) antibody titre (35-day-SRBC test) performance; the 1% amaranth treatment had a higher antibody titre than the 2% treatment.

Among the taste and sensory traits of meat, only meat aroma was affected ($p < 0.01$) by treatment. A difference was observed between treatments (1% and 2% amaranth) compared to the control treatment. Also, the results of the contrast of the control group compared to the amaranth treatments, the linear equation (y = 0.6x + 6.8667, the coefficient of determination 0.35, and the quadratic for the meat aroma trait were significant ($p < 0.05$).

Table 6. Results of parameters related to the immune system and sensory and taste attributes of broiler chickens' breast meat

Treatments	Antibody Titre (35-day- SRBC test)	Antibody Titre (42-day- SRBC test)	Pancreas weight (g)	Spleen weight (g)	Fabricius weight (g)	Meat aroma	Meat taste	Meat smell	Meat crispy	Meat color	Desirability and acceptance of meat
T_1 (control)	2.400^{ab}	6.400	5.400	2.300	1.660	6.40^{b}	7.40	8.40	8.00	7.20	7.80
T_2	3.600^{a}	4.400	5.040	2.540	1.740	8.40^{a}	7.60	7.00	7.80	8.20	7.60
T_3	1.200^{b}	4.800	5.300	2.680	2.240	7.60^{a}	7.60	8.00	7.80	7.60	8.20
SEM	0.432	0.959	0.353	0.257	0.428	0.306	0.476	0.392	0.440	0.424	0.383
p-value	0.007	0.330	0.763	0.586	0.597	0.002	0.943	0.068	0.934	0.282	0.546
Control Vs. Amaranth	0.999	0.151	0.605	0.344	0.541	0.001	0.738	0.085	0.717	0.203	0.835
Linear	0.073	0.261	0.845	0.316	0.357	0.017	0.772	0.484	0.753	0.518	0.474
Quadratic	0.005	0.327	0.488	0.876	0.696	0.003	0.867	0.028	0.856	0.150	0.411

* Means with the same letter for each column are not significantly different
* T_1: control group: 0% amaranth, T_2: 1% amaranth, T_3: 2% amaranth

The significant quadratic equation of antibody titre (35-day-SRBC test) was as follows. Letter y is the target trait, and letter x is the amaranth level.

Antibody Titre (35 day-SRBC test) = $-1.8x^2 + 3x + 2.4$ min = 0.83

Table 7 shows the fatty acid profiles in broiler breast meat. Given that only one breast sample from each treatment was analyzed, accurate comparisons and statistical analysis are not possible. However, in general, consumption of amaranth led to numerically higher indicators related to improving the quality of breast meat (e.g., a decrease in SFA, an increase in MUFA, PUFA, an increase in UFA, a decrease in the ratio of omega-6 to omega-3 fatty acids, and a decrease in the atherogenic index). In future studies, a detailed statistical analysis of multiple breast meat samples is needed.

Humans consume broiler meat. Any effort to improve the quantity and quality of chicken meat can have a direct impact on the health of the human consumer. Therefore, this study was conducted to assess the feasibility of using amaranth in feeding broiler chickens to increase their health, as well as the quantity and quality of their meat products. The findings of the present study showed that the use of amaranth significantly improved body weight and FCR in the starter period and that similar, not reduced, performance was obtained in the other periods and in the entire feeding period compared to the control group. These results were consistent with the first study on amaranth feeding in chickens (Connor et al., 1980), which obtained similar performance in control versus amaranth-fed chickens. The observed improved growth performance after feeding amaranth to chickens (Laovoravit et al., 1986) was in line with some of the results of this study. The reason for some differences in other feeding periods can be attributed to the level of feed consumption, the type of amaranth used, and its processing status. Other researchers (Tillman & Waldroup, 1986) also observed weight gain and improved FCR in broilers after feeding up to 20% amaranth, but this was not observed at higher levels and in raw form, which was consistent with the general results of this study. The reason for some differences was attributed to the type of processing and levels used. When feeding up to 40% extruded amaranth, similar performance between amaranth-fed and control broilers was observed (Tillman & Waldroup, 1988), consistent with the results of the present study.

After feeding different forms of raw amaranth, similar performance in amaranth-fed and control broilers was observed (Acar et al., 1988), consistent with the general results of this study; the minor differences were due to the type of amaranth and levels used. Raw amaranth grain fed at levels of 0, 20, 40, and 60% reduced feed intake and weight gain in broiler chickens, and subsequently impaired FCR (Ravindran et al., 1996), but this impairment was overcome by heat treatment. It is hypothesized that amaranth, due to its high fiber content compared to other grains and the presence of a number of anti-nutritional compounds at high levels of raw consumption, may not improve performance. Given the low levels of amaranth consumption in the present study, this hypothesis is consistent with the results of this study. In the present study, high levels of consumption were avoided, and even phytase enzyme was used in the diet formulation. No performance loss was observed in the starter period, but also in the entire strengthening period, and at the end of the period.

We examined two varieties of amaranth (*A. hypochondriacus* and *A. cruentus*) in the form of grain for broiler chickens in raw and processed forms and found that 20% of raw and autoclaved amaranth did not differ significantly from the control group in terms of performance, which was consistent with the present study despite the species difference. The use of up to 7% amaranth as a substitute for meat and bone meal waste resulted in a similar performance to the control group (Rouckova et al, 2004),

consistent with the general findings of the present study. A decrease in performance was observed when supplementing broiler chicken diets with *A. caudatus* grains (Longato et al., 2017), which was in contrast to the results of this study; the reason for this difference may be attributed to the levels of feed consumption and the species of grain amaranth used.

Feeding broiler chickens a diet containing 8% raw amaranth resulted in reduced feed intake and body weight (Orczewska-Dudek et al., 2018), which was in contrast to the results of this study; however, no negative effect on FCR was observed after feeding amaranth, which is in agreement with the present study. To determine the optimal levels of *A. hybridus* in pelleted diets for broiler chickens, a multi-indicator decision-making method and production index were used to obtain an estimated value of 2.0% and 1.57% amaranth, respectively, for the feeding of broiler chickens (Alizadeh-Ghamsari et al., 2021). These researchers hypothesized that using raw amaranth in the pellet diet of broilers up to 2% can attain the best performance, and levels above 2% require processing and evaluation, as well as consideration of other parameters related to bird health, which was consistent with the results of this study.

Table 7. Fatty acid profile of broiler breast meat obtained at 42 days of age

Items	Treatments		
	T_1	T_2	T_3
Caprylic acid (C8:0)	0.85	0.08	0.02
Capric acid (C10:0)	0.98	0.07	0.01
Lauric acid (C12:0)	8.95	0.61	0.15
Myristic acid (C14:0)	4.07	0.75	0.58
Pentadecanoic acid (C15:0)	-	0.08	0.07
Palmitic acid (C16:0)	22.44	24.30	24.40
Margaric acid (C17:0)	0.14	0.12	0.11
Stearic acid (C18:0)	6.83	7.05	6.42
Arachidic acid (C20:0)	0.26	0.26	0.25
Behenic acid (C22:0)	0.22	0.44	0.34
Total SFA	44.74	33.76	32.35
Myristoleic acid (C14:1)	-	0.12	0.15
Palmitoleic acid (C16:1)	2.50	3.62	4.90
Elaidic acid (C18:1t)	0.10	0.12	0.13
Oleic acid (C18:1c)	27	30.38	34.73
Gondoic acid (C20:1)	0.13	0.08	0.07
Erucic acid (C22:1)	-	0.03	-
Total MUFA	56.86	64.81	74.78
Linoleic acid (C18:2c)	22.43	27.33	24.13
Linolelaidic acid (C18:2c)	22.43	27.33	24.13
Cis-11,14-Eicosadienoic acid (C20:2)	0.16	0.35	-
Arachidonic acid (C20:4)	-	0.11	0.04
Cervonic acid (22:6)	-	0.28	0.15
Total PUFA, n-6	45.02	55.40	48.45
Dihomo-γ-linolenic acid (C20:3)	0.88	1.87	1.19
Linolenic acid (18:3)	1.36	1.81	1.69
Total PUFA, n-3	2.24	3.68	2.88
Total PUFA, n-6/Total PUFA, n-3	20.09	15.05	16.82
Other	0.01	0.12	0.47
UFA	101.89	120.33	123.70
UFA/SFA	2.27	3.56	3.82
Atherogenic index (AI)	16.49	3.19	2.51
Thrombogenic index (TI)	3820.70	4199.60	4729.11
Hypocholesterolemia index (HI)	1.94	2.46	2.48
Hypocholesterolemic / Hypercholesterolemic	3.84	4.80	4.95

PUFA: polyunsaturated fatty acids, MUFA: monounsaturated fatty acids, UFA: unsaturated fatty acids, SFA: Saturated fatty acids, T1: control group: 0% amaranth, T2: 1% amaranth, T3: 2% amaranth.

A decrease in performance after feeding *A. spinosus* at levels above 20% was observed (Gebeş et al., 2024), which can be attributed to the difference in processing methods and plant species. However, it is hypothesized that the species investigated in the present study can be considered as an enriching and economical plant for broiler chickens at low consumption levels without having a negative impact on performance. On the other hand, it can meet the minimum requirements of the bird with minimal metabolic pressure on the bird and the environment, without the need for thermal processing and thermal energy consumption.

The results of the present study indicated no significant decrease or negative effect on carcass component weight after feeding amaranth. These results were in contrast to the results of other researchers (Orczewska-Dudek et al., 2018; Gebeş et al., 2024) who observed a linear increase in carcass performance, especially for breast meat, after feeding amaranth. The reason for this was attributed to the use of higher levels of amaranth and increased dietary protein and increased feed intake, which recommended the need for further studies in these areas. Results of one study (Alizadeh-Ghamsari et al., 2021) showed that including 6% amaranth (*A. hybridus*) in pelleted diets for broilers reduced carcass performance, but at levels of 2% or 4% amaranth, there was no difference compared to the control group, a finding consistent with the general results of the present study.

Given the high fiber content of amaranth, it is hypothesized that increasing the levels of amaranth consumption in the diet would increase gizzard and liver weight in poultry (Orczewska-Dudek et al., 2018; Gebeş et al., 2024), but this was not observed in the present study due to the selected optimal levels of difficulty and negative effects. No negative effects on the gizzard up to a 6% amaranth feeding level were observed (Alizadeh-Ghamsari, 2021), which was consistent with the results of the present study.

With amaranth consumption levels of 10, 20, 30, 40, and 50%, a decreasing trend in fat pad weight, heart weight, gizzard weight, and liver weight were observed with increasing levels of amaranth (Tillman & Waldroup, 1988), indicating that lower levels of amaranth consumption by broilers, such as in the present study, were selected to prevent this trend.

For blood biochemical parameters, feeding grain amaranth resulted in an improvement and a promising trend for many items related to bird health. The decrease in blood glucose, cholesterol, LDL, Atherogenic Index, ALT, AST, and MDA, as well as the parallel increase in HDL, uric acid, albumin, total protein, phosphorus, Fe, Ca, and TAC after feeding amaranth was quite significant.

In general, amaranth has the potential to improve the biochemical and antioxidant status of the blood because it contains several powerful cholesterol-lowering agents, such as crude fiber, squalene, phytosterols, tocopherols, tocotrienols, and sterols (Tang & Tsao, 2017). Decreased blood glucose was observed when feeding raw amaranth to laying hens, consistent with the present investigation. A review of the scientific literature has shown that amaranth grains contain effective compounds that can prevent fructose-induced obesity and diabetes. In addition to specific bioactive compounds, some known compounds (such as amaranth tocotrienols) have antioxidant effects and diabetes-reducing properties of their own (Tang & Tsao, 2017). Amaranth grains contain dipeptidyl aminopeptidase (DPP IV) inhibitor, a key enzyme responsible for inactivating gastric inhibitory polypeptide (GIP) and glucagon-like peptide-1 (GLP-1), which is an insulin-releasing hormone for endocrine cells in the gut (O'Harte et al., 1999). In addition, a unique compound specific to amaranth called 20-hydroxyecdysone (20HE) is a bioactive compound shown to significantly reduce blood glucose and lead to anti-diabetic properties (Tang & Tsao, 2017).

In general, the reduction in blood glucose in this study can be attributed to the presence of such compounds, which indicates the power of amaranth, even at low levels, for monogastric animals such as poultry. These findings can be generalized even for human studies and have high study value. In addition, some known and specific compounds unique to amaranth grains can limit the rate of production of the hepatic enzyme HMG-CoA reductase, which can lead to a decrease in blood cholesterol. The presence of high fiber in amaranth leads to its cholesterol-lowering effects by binding to cholesterol and excreting it. The presence of high linoleic acid in amaranth grains also plays an important role in the excretion of bile acids and in reducing cholesterol. Also, an exclusive compound of amaranth grains is squalene, which, if supplied to the body through nutrition, can go to the liver with the help of chylomicrons and be used to make steroids and bile acids. In parallel with these events, bile acids (cholic and deoxycholic) synthesized from squalene are produced in liver cells, which can combine with glycine and taurine to produce bile salts, which can lead to a decrease in cholesterol by increasing bile secretion turnover (Kianfar et al., 2023).

Supplementing the diet of chickens with *A. hypochondriacus* and *A. cruentus* resulted in a 10-30% reduction in blood cholesterol (Qureshi et al., 1996). The LDL levels were also reduced by 7-70%. In general, a 10-18% reduction in the enzymatic activity of cholesterol 7-alpha hydroxylase and 3-hydroxy-3-methylglutaryl coenzyme A reductase in the liver compared to other experimental groups was the reason for the significant difference in blood cholesterol levels of experimental birds. Activation of the enzyme 7-alpha hydroxylase leads to the formation of bile acids from cholesterol and increases the catabolism and excretion of cholesterol. The results of that study were in agreement with the present study.

A study with hamsters (Mendonça et al., 2009) confirmed the cholesterol-lowering effects of amaranth grains and attributed the reason to the presence of peptides that limit the production of the HMG-CoA reductase enzyme. Additional studies and a review of the scientific literature showed that a special phytochemical compound in amaranth grains called 20-hydroxyacetidone (20HE), in an amount of 148-484 mg, in addition to reducing blood glucose, can enhance anti-obesity effects such as reducing cholesterol and LDL (Tang & Tsao, 2017). Also, the possibility of the presence of tocopherols in amaranth can play an effective role in reducing the synthesis of cholesterol, low-density lipoprotein, and lipoprotein lipase enzyme, and ultimately regulating and reducing cholesterol in the blood (Qureshi et al., 1996; Tang & Tsao, 2017). The presence of eight isomers of vitamin E in amaranth was stated as the reason for the 30% and 70% reduction in cholesterol and LDL in chickens (Hood, 1998). Increasing the level of amaranth grains did not have any negative effect on the level of red blood cells, hemoglobin and iron content in laying hens (Króliczewska et al., 2008), but in parallel, a decrease in blood lipid parameters, including a decrease in cholesterol and LDL and a change in AST and ALT activity, was observed compared to the control group, which was consistent with the results of this study. No difference in blood parameters such as iron, AST and ALT, total protein, albumin, total cholesterol, HDL and triglycerides was observed when feeding extruded amaranth to laying hens at levels of 5 and 10% (Popiela et al., 2013), which was in contrast to the present study; the reason for this was the processing mentioned in the study, which did not have a significant effect on the aforementioned parameters. In the form of raw amaranth in this study, the effect was noticeable for iron, calcium, albumin, protein, and blood uric acid, and this is in line with the improvement of bird health. Glucose, AST, and ALT levels decreased with increasing levels of extruded amaranth grains (Popiela et al., 2013), consistent with the present report.

Abundant amounts of phytosterols in the grain oil content of *A. hybridus* species were reported (Marcone et al., 2004), which justifies its cholesterol-lowering power. A significant decrease in serum lipid peroxidation levels was observed after feeding 0, 5, or 10% *A. caudatus* grains to female broilers (Longato et al., 2017), consistent with most of the results of the present investigation. By supplementing broiler chickens' diets with *A. caudatus* grain, the oxidative status and total blood antioxidant capacity were significantly increased (Longato et al., 2017). Increasing grain amaranth consumption levels (0, 2, 5, or 10%) by laying hens resulted in reductions in blood ALT and AST activity compared to the control group (Króliczewska et al., 2008). Considering that many poultry are raised in managed environments and are likely to face various types of stresses, the use of valuable antioxidant food sources such as amaranth in their diet can be considered a reliable and low-risk mitigation solution (Hosseintabar-Ghasemabad et al., 2024).

A review of the scientific literature has shown that amaranth bioactive compounds have always been effective in strengthening the blood antioxidant system, and one of the prominent amaranth compounds is squalene. Squalene is essentially an antioxidant that, due to its high electron exchange capacity and without being exposed to molecular disorder, can lead to improved macrophage function and strengthen the immune and antioxidant system. Since squalane can provide biological protection against oxidative stress during phagocytosis, it can protect the biological membrane of immune cells (Seidavi et al., 2023). Squalane has a similar structure to other isoprenoids such as beta-carotene, lycopene, vitamin A, vitamin E, and coenzyme Q_{10} (ubiquinone), which can produce superior and effective antioxidant effects like the aforementioned active compounds in the body. Güneş (2013) believes that because squalane is a non-polar substance that tends to bind to non-ionized substances, it can help in the removal of harmful substances and detoxification of the body.

The unique compounds of amaranthine and isomaranthine in amaranth grains can affect tocopherol catabolism, leading to their accumulation in serum and tissues, which in turn can play an important role in improving the antioxidant activity of the blood (Tsao & Tang, 2017). Nitric oxide (NO) is one of the most important signaling molecules against oxidative stress that has the potential to reduce various types of damage caused by reactive oxygen species. Amaranth grains have very strong activity against superoxide radicals, can have a positive effect on macrophages, and help modulate immunity by producing sufficient NO in the body cells (Tang & Tsao, 2017). Selenium and betacyanin present in amaranth grains also have anti-inflammatory properties and contribute to the antioxidant system of blood cells by inhibiting NFkB in the cell nucleus (Tang & Tsao, 2017). Even recently, despite the potential negative effects of phytate in poultry nutrition, the class of phytates (phytate-containing compounds) present in raw amaranth can play a colorful role by inhibiting the production of hydroxyl radicals and normalizing cell homeostasis, leading to increased and improved antioxidant activity to enhance consumer health; low levels of amaranth consumption can help to provide limited phytate in the diet for consumer health (Tang & Tsao, 2017).

Dietary manipulation can affect broiler productivity (Ghane-Khoshkebijari et al., 2024; Shaban et al., 2024). In the present study, the results were promising, as no adverse effects were observed after feeding amaranth. Similarly, Longato et al. (2017) reported no negative effects on meat quality, which is consistent with our findings. However, Gebeş et al. (2024) noted an increase in meat pH following amaranth supplementation, a parameter not assessed in the present work. Other sensory and taste traits were unaffected, with the exception of meat aroma, which may have been influenced by bioactive compounds present in raw amaranth. Gebeş et al. (2024) have suggested that meat quality and sensory characteristics are closely related to the antioxidant status of poultry. Given that dietary amaranth improved blood antioxidant status in this study, a corresponding enhancement in overall body antioxidant capacity can be inferred. While no negative effects on meat quality were observed, the potential of amaranth to improve sensory attributes and meat healthfulness warrants future research.

The results of this study show that dietary supplementation with *Amaranthus hybridus chlorostachys* at levels up to 2% in broiler diets did not negatively affect animal performance or carcass yield, while improving several health-related meat attributes, such as reduced cholesterol content and increased antioxidant capacity. These changes are not only relevant to animal husbandry economics but also provide public health benefits for poultry consumers. Specifically, the observed reductions in blood cholesterol and improvements in meat antioxidant status observed in broilers suggest potential implications for human nutrition, as such meat profiles could contribute to decreased dietary cholesterol intake and increased intake of natural antioxidants. These changes may play a role in the prevention of chronic diseases, including cardiovascular and metabolic syndrome, aligning with the broader goals of health promotion and disease prevention. Additionally, promoting the use of drought-resistant and nutrient-rich crops like amaranth as sustainable feed ingredients supports food system resilience, environmental stewardship, and community health, thereby addressing the human-environment interface central to health promotion.

The use of *Amaranthus hybridus* grain in animal diets demonstrates several health-promoting effects for both animals and, by extension, humans. These include hypocholesterolemic properties, improvements in antioxidant status, and a potential reduction in risk factors for chronic non-communicable diseases due to the bioactive compounds present in the grain, such as dietary fiber, squalene, and polyphenols. In contrast, the unauthorized synthetic food colorant "Amaranth" (E123) is banned in multiple countries due to links with adverse health outcomes, including potential genotoxic and carcinogenic risks. If there is ambiguity regarding the type of amaranth referenced, this could undermine public health messaging

and food safety efforts. Therefore, rigorous clarification and public education must distinguish between the botanical ingredient and the synthetic dye. From a public health and regulatory perspective, carefully controlled use of *Amaranthus hybridus* grain in animal feed represents a potential avenue for increasing the nutritional value and safety of animal-source foods. In contrast, the continued prohibition of the synthetic colorant is justified by precautionary principles in food hygiene and public health protection.

By highlighting the scientifically documented health-promoting effects of botanical amaranth grain in the animal model-distinct from the risks associated with synthetic amaranth colorant-the current manuscript strongly supports the relevance of our work to food safety, public health interventions, and environmental health, thus fitting well within the human health promotion topics.

3.1 Implications, limitations, and entry points for future research to show future

The importance and necessity of conducting this study lie in the growing demand for sustainable, health-promoting, and cost-effective alternatives to conventional feed ingredients in the poultry industry. With increasing concerns over the environmental impact, economic volatility of feed resources, and consumer preference for functional and enriched animal products, the exploration of *Amaranthus hybridus chlorostachys* as a natural dietary supplement offers a promising solution. This plant is nutritionally rich and contains bioactive compounds such as squalene, polyphenols, and unsaturated fatty acids, which may enhance broiler health, immunity, meat quality, and antioxidant status. Despite its potential, limited empirical data exist on the optimal inclusion levels and holistic effects of raw amaranth in broiler diets, particularly in its unprocessed form. Thus, this study is essential to fill the knowledge gap and provide evidence-based insights into the safe and beneficial incorporation of amaranth in poultry nutrition, supporting both industry innovation and public health objectives.

Based on our limitations, we can demonstrate that research based on performance and product evaluation should be avoided, and further mechanistic studies should be conducted to understand how the compounds extracted from medicinal plants affect livestock and poultry species. The advantages of this work included site of action, effective doses, *in vivo* metabolism (by the animal and microflora in the digestive tract), *in vivo* digestibility, effect of variety type, plant growth conditions, processing, and storage on active compounds or secondary metabolites of the plant.

When these cases occur, the use of medicinal plants and their extracts can be applied. Moreover, standardization of the effects of herbal products using bioactive compound encapsulation techniques is a suitable method for the production of these herbal products. Increasing the availability and absorption of active compounds and coated extracts can be useful in formulating these compounds. Also, standardization of the extraction method is to extract the active ingredients of medicinal plants and to develop a uniform protocol and procedure according to the type of medicinal plant variety. Meanwhile, the use of herbal digraphides (essential oils and extracts) and the investigation of the antioxidant, antibacterial, and antiviral effects of these natural compounds are needed in further research. The use of the effects of medicinal plants on the quality and quantity of meat and sensory evaluation should be comprehensively investigated. Investigation of the effects of herbal compounds used in this study on gene expression (such as interleukin-6 and interferon gamma, mucin, etc.), and also the study of the effects of other medicinal plants on the expression of cytokine genes as indicators of the humoral and cellular immune system. Finally, the effects of medicinal plants and their extracted substances on immunological properties (such as the investigation of the anticoccidial activity of medicinal plants, their extracts, and essential oils, and the reduction of necrotic intestinal swelling when using these compounds) should be studied.

4. Conclusion

In the present study, amaranth showed good potential as a feed source for enrichment in the diet of broiler chickens to help improve health-promoting items, especially blood biochemical parameters. In addition, the use of amaranth with the lowest level of selection and without metabolic pressure on the animal, as well as to help the environment (no heat processing) to produce optimally and easily, was able to improve the performance of chickens in the starter period and also improve the quality, sensory, and taste of meat. In parallel, without any negative effects on other performance traits, carcass characteristics, immune system, and some blood parameters, it offers promising potential and impressive achievements for the successful prospect of using amaranth in feeding programs. This finding can promote health in humans who consume broiler meat. The use of higher-quality products resulting from the achievements of this research can lead to improving the health of society.

Authors' Contributions

Mahmoud Saei: Data curation; Formal analysis; Investigation; Writing-original draft. **Alireza Seidavi:** Conceptualization; Project administration; Supervision; Validation; Writing-review & editing. **Mehrdad Bouyeh:** Conceptualization; Project administration; Validation; Writing-review & editing.

Funding

Financial support from the Rasht Branch, Islamic Azad University (grant number 6298.1) is gratefully acknowledged.

Conflicts of Interest

The authors declare that they have no conflict of interest.

Acknowledgments

This article is based on the PhD thesis of the first author at Ra.C., Islamic Azad University, Rasht, Iran. The authors would like to express their gratitude to MSc. GH. Hosseintabar, Director of Darvash Giah Khazar Medicinal Herbs Complex, for providing the plant-based raw materials and for his valuable scientific and technical support. The authors also sincerely thank the management and staff of the R&D unit of Sepid Makian Company, Viromed, and Tekno Azma Laboratories for their cooperation and technical support in this project.

Ethical considerations

The use and care of birds in this experiment were approved by the Ethics Committee of the Rasht Branch, Islamic Azad University, Rasht, Iran. (Code: IR.IAU.RASHT.REC.1402.021). All experimental procedures were conducted in accordance with the approved protocols, and efforts were made to minimize the number of birds used.

Using artificial intelligence

No artificial intelligence (AI) techniques were used in this study.

References

Alizadeh-Ghamsari, A. H., Hosseini, S. A., Soleymani, M. R., & Nahavandi, R. (2021). Performance, intestinal histomorphology, and blood variables of broilers fed amaranth grain in pellet diet. *Tropical Animal Science Journal*, *44*(1), 71-78.

Amirdahri, S., Janmohammadi, H., Taghizadeh, A., Olyayee, M., Hosseintabar-Ghasemabad, B., Seidavi, A., & Vlčková, R. (2023). Dietary leucine deficiency alters performance, body composition, intestine microbiota, and immune response of female Cobb broilers. *Animal Production Science*, *63*(17), 1750-1758.

Attia, Y. A., Al-Harthi, M. A., Al-Sagan, A. A., Alqurashi, A. D., Korish, M. A., Abdulsalam, N. M., . . . & Bovera, F. (2022). Dietary supplementation with different ω-6 to ω-3 fatty acid ratios affects the sustainability of performance, egg quality, fatty acid profile, immunity and egg health indices of laying hens. *Agriculture*, *12*(10), 1712.

Azri, A., Aydi, S. S., Aydi, S., Debouba, M., Bouajila, J., Cerny, M., . . . & Merah, O. (2025). Nutritional and bioactive lipid composition of *Amaranthus* seeds grown in varied agro-climatic conditions in France. *Agronomy*, *15*(3), 672.

Bejosano, F. P., & Corke, H. (1998). Protein quality evaluation of *Amaranthus* whole meal flours and protein concentrates. *Journal of the Science of Food and Agriculture, 76*(1), 100-106.

Connor, J. K., Gartner, R. J. W., Runge, B. M., & Amos, R. N. (1980). *Amaranthus edulis*: An ancient food source re-examined. *Australian Journal of Experimental Agriculture and Animal Husbandry*, *20*(103), 156-161.

Food and Agriculture Organization of the United Nations (FAO). (2023). *Amaranth production statistics.* https://www.fao.org/statistics/en/

Gebeş, E. S., Yalçın, S., Ramay, M. S., Ünal, A., & Filikçi, K. (2024). Suitability of raw and heat-treated *Amaranthus spinosus* in broiler diets: Effects on growth performance, meat antioxidant capacity, haemato-biochemical parameters, intestinal histomorphometry, and cecal volatile fatty acid profile. *Tropical Animal Health and Production*, *56*(7), 238-242.

Ghane-Khoshkebijari, F., Seidavi, A., & Bouyeh, M. (2024). The impact of *In Ovo injection* of organic manganese on broiler breeder hen egg hatchability and progeny broilers productivity. *Journal of Human Environment and Health Promotion*, *10*(4), 229-237.

Güneş, F. E. (2013). Medical use of squalene as a natural antioxidant. *Journal of Marmara University Institute of Health Sciences*, *3*(4), 220-228.

Hood, R. L. (1998). *Tocotrienols in metabolism.* Phytochemicals: A New Paradigm.

Hosseintabar-Ghasemabad, B., Di Rosa, A. R., Janmohammadi, H., Slozhenkina, M. I., Gorlov, I. F., Mosolov, A. A., . . . & Phillips, C. J. C. (2024). The potential of amaranth grain for feeding to poultry. *World's Poultry Science Journal*, *80*(2), 481-509.

Janmohammadi, H., Hosseintabar-Ghasemabad, B., Amirdahri, S., Gorlov, I. F., Vladimirovna, K. E., Slozhenkina, M. I., . . . & Phillips, C. J. C. (2022). The energy value for broiler chickens of heat-treated and untreated amaranth grain, with and without enzyme addition. *Agriculture*, *12*(11), 1810.

Kianfar, R., Di Rosa, A. R., Divari, N., Janmohammadi, H., Hosseintabar-Ghasemabad, B., Oteri, M., . . . & Seidavi, A. (2023). A comparison of the effects of raw and processed amaranth grain on laying hens′ performance, egg physicochemical properties, blood biochemistry and egg fatty acids. *Animals*, *13*(8), 1394.

Króliczewska, B., Zawadzki, W., Bartkowiak, A., & Skiba, T. (2008). The level of selected blood indicators of laying hens fed with addition of amaranth grain. *Electronic Journal of Polish Agricultural Universities*, *11*(2), 18.

Laovoravit, N., Kratzer, F. H., & Becker, R. (1986). The nutritional value of amaranth for feeding chickens. *Poultry Science*, *65*(7), 1365-1370.

Longato, E., Meineri, G., & Peiretti, P. G. (2017). The effect of *Amaranthus caudatus* supplementation to diets containing linseed oil on oxidative status, blood serum metabolites, growth performance and meat quality characteristics in broilers. *Animal Science Papers and Reports, 35*(1), 71-86.

Manual, R. B. (2012). *Ross 308 broiler nutrition manual.* Ross Broiler Ltd: Newbridge, Midlothian, UK.

Marcone, M. F., Kakuda, Y., & Yada, R. Y. (2004). Amaranth as a rich dietary source of β-sitosterol and other phytosterols. *Plant Foods for Human Nutrition*, *58*(3), 207-211.

Mendonça, S., Saldiva, P. H., Cruz, R. J., & Arêas, J. A. (2009). Amaranth protein presents cholesterol-lowering effect. *Food Chemistry*, *116*(3), 738-742.

Mozaffarian, V. (1996). *A dictionary of Iranian plant names*. Farhang Moaser.

Nwadinigwe, A. O., Amadi, C. C., Amujiri, A. N., Nwadinigwe, C. C., & Ozokolie, C. B. (2019). Effects of pharmaceutical effluents on germination, growth and development of *Amaranthus hybridus* L. *Nigerian Journal of Botany*, *32*(2), 1-11.

O'Harte, F. P., Mooney, M. H., & Flatt, P. R. (1999). NH_2-terminally modified gastric inhibitory polypeptide exhibits amino-peptidase resistance and enhanced antihyperglycemic activity. *Diabetes*, *48*(4), 758-765.

Orczewska-Dudek, S., Pietras, M., & Nowak, J. (2018). The effect of amaranth seeds, sea buckthorn pomace and black chokeberry pomace in feed mixtures for broiler chickens on productive performance, carcass characteristics and selected indicators of meat quality. *Annals of Animal Science*, *18*(2), 501-523.

Patro, A., & Chowdhury, A. (2025). Pseudocereals: Nutritional powerhouse for a healthier future. *Vigyan Varta an International E-Magazine for Science Enthusiasts, 6*(3), 185-188.

Phillips, C. J., Hosseintabar-Ghasemabad, B., Gorlov, I. F., Slozhenkina, M. I., Mosolov, A. A., & Seidavi, A. (2023). Immunomodulatory effects of natural feed additives for meat chickens. *Life*, *13*(6), 1287.

Popiela, E., Króliczewska, B., Zawadzki, W., Opaliński, S., & Skiba, T. (2013). Effect of extruded amaranth grains on performance, egg traits, fatty acids composition, and selected blood characteristics of laying hens. *Livestock Science*, *155*(2-3), 308-315.

Qureshi, A. A., Lehmann, J. W., & Peterson, D. M. (1996). Amaranth and its oil inhibit cholesterol biosynthesis in 6-week-old female chickens. *The Journal of Nutrition*, *126*(8), 1972-1978.

Ravindran, V., Hood, R. L., Gill, R. J., Kneale, C. R., & Bryden, W. L. (1996). Nutritional evaluation of grain amaranth (*Amaranthus hypochondriacus*) in broiler diets. *Animal Feed Science and Technology*, *63*(1-4), 323-331.

Rouckova, J., Trckova, M., & Herzig, I. (2004). The use of amaranth grain in diets for broiler chickens and its effect on performance and selected biochemical indicators. *Czech Journal of Animal Science, 49*(12), 532-541.

Seidavi, A., Hosseintabar-Ghasemabad, B., Gorlov, I. F., Slozhenkina, M. I., Mosolov, A. A., Oteri, M., . . . & Luigi, L. (2023). Feed additives and future perspectives. In G. Arsenos, & L. Giannenas, *Sustainable use of feed additives in livestock: Novel ways for animal production* (pp. 929-961). Springer International Publishing.

Shaban, E., Bouyeh, M., & Seidavi, A. (2024). The Impact of reduced licorice levels on performance, carcass characteristics, blood constituents, immunity, intestinal microflora, intestinal morphology, and breast muscle fatty acid profile in broilers. *Journal of Human Environment and Health Promotion*, *10*(3), 161-169.

Tang, Y., & Tsao, R. (2017). Phytochemicals in quinoa and amaranth grains and their antioxidant, anti-inflammatory, and potential health beneficial effects: A review. *Molecular Nutrition and Food Research*, *61*(7), 1600767.

Tillman, P. B., & Waldroup, P. W. (1986). Processing grain amaranth for use in broiler diets. *Poultry Science*, *65*(10), 1960-1964.

Tillman, P. B., & Waldroup, P. W. (1988). Performance and yields of broilers fed extruded grain amaranth and grown to market weight. *Poultry Science*, *67*(5), 743-749.