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Research Article

The Effect of Replacing Sorghum Grains with Corn Along with Phytase and NSP Enzymes on Yield and Blood Parameters of Broilers

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ABSTRACT
This study explores the impact of replacing grain sorghum with corn, supplemented with phytase and NSP enzymes, on broiler chicken performance and blood parameters. Various sorghum levels (0%, 5%, and 10%) and two enzyme concentrations (0% and 0.1%) were incorporated into broiler feed. Blood samples were collected at 42 days, measuring blood serum parameters. Throughout the trial, feed consumption, daily weight gain, food conversion ratio, and carcass characteristics were assessed. Experimental treatments included a control diet (no sorghum, no enzyme), and combinations of sorghum and enzymes. Employing a 2x3 factorial method with a completely random design, the study utilized 6 treatments and 3 repetitions, involving 270 Ross 308 broiler chickens. Results revealed significant differences in sorghum's main effects on feed consumption in the first and sixth weeks, and enzyme effects only in the last week ($p<0.05$). Live weight gain showed significance in sorghum's main effects during the fifth and sixth weeks ($p<0.05$). Food conversion ratio differences were observed in sorghum's main effects in the fifth week ($p<0.05$), with the diet without sorghum displaying the highest ratio. Carcass traits exhibited significant differences in sorghum's main effects ($p<0.05$). Blood tests showed significant differences in cholesterol, HDL, and LDL levels due to sorghum ($p<0.05$). Overall, while the addition of sorghum and enzymes did not consistently impact feed consumption, live weight, and carcass traits, a 10% sorghum level positively influenced food conversion ratio in certain weeks of the experiment.

Keywords: *Broiler; Enzyme; NSP; Phytase; Sorghum.*

Introduction

Due to the increasing growth of the poultry industry, Poultry producers are thinking of ways to produce protein, including white meat, in the shortest possible time with lower costs and with the maximum possible growth. Produce in broilers. Improving and increasing the production parameters in broilers is one of the most important goals of the poultry industry in the whole world. Today, various breeding techniques, medicinal substances, and natural growth supplements have been presented to achieve these goals [1]. Many different factors could affect the growth and production of animals including genetics, nutrition, and environment [2], [3], [4]. In the past, the use of growth-promoting antibiotics effectively reduced the need to use enzymes. But in 2006, the European Union banned the use of all growth-promoting antibiotics used in animal feed. In the United States, the Food and Drug Administration has banned the use of most growth-promoting antibiotics, so researchers are looking for additives to replace antibiotics. Enzymes are a group of proteins that can act as a very strong biological catalyst. be used in the feed industry. Considering that enzymes are not toxic or have very little toxicity, and also many of them can maintain their biological activity in the range of pH and temperature, enzymes can be good substitutes for some additives in the market [5].

In most broiler chicken breeding units, corn is used as one of the main components in the diet, and due to the high need and lack of corn and its high price in the market, as well as low imports in Iran, many researchers are trying to replace it with other sources. Due to the structural similarity of sorghum with corn, there is a need for a lot of research in the field of using sorghum (*Sorghum bicolor* L. Moench) instead of corn. In terms of importance, sorghum ranks fifth among cereals in the world after wheat, rice, corn, and barley. Sorghum is tolerant to drought and relatively resistant to pests and diseases. The amount of sorghum protein varies from 8 to 16 percent, and commercial cultivars have 10 to 13 percent protein. The amounts of lysine, methionine, crude fiber, ash, and phosphorus in sorghum are similar to corn. The sorghum grains have a substance called tannin, less than two percent of which is considered a positive factor in nutrition. Because sorghum has a significant amount of phytate-phosphorus and soluble non-starch polysaccharide (NSP), it is an ideal food that has a lot of potential in chicken diets. Research shows that the amount of (NSP) of the desired raw materials along with microbial enzymes is reduced and food quality is improved [6], [7]. In general, using sorghum can be economically justifiable and when it comes to animal husbandry considering commercial productivity is very crucial [8].

Today, commercial additive products, including microbial phytase enzymes, are considered a part of the main components of poultry feed and are considered an effective method to increase the use of phosphorus phytate in poultry nutrition and reduce phosphorus excretion and environmental pollution [9]. There is a large amount of phytic acid in diets based on plant sources that are fed to domestic animals. Digestion of phytic acid in the digestive system of monogastric animals is very low, because of this, phosphorus phytate is excreted and causes pollution of the environment and freshwater resources. In addition, low phosphorus digestion increases production costs, because other sources of phosphorus must be used to adjust dietary phosphorus.

In addition, phytate has a negative effect on mineral absorption and protein digestibility. One of the tools for developing and perfecting the technology of producing phytase enzyme from microbes with the help of genetic engineering and adding it to the diet of domestic animals has been to increase the digestibility of phosphorus phytate. Today, phytase enzyme is used as an additive to improve the bioavailability of phosphorus in poultry feed [1]. In any case, when using phytase in the diet, attention should be paid to its effect on the release of phosphorus and some other mineral elements, especially calcium, because the balance of calcium and phosphorus in the diet, in addition to affecting the animal, is also involved in the excretion of phosphorus and environmental pollution. Adding phytase to the diet leads to an increase in the bioavailability of amino acids and also to an increase in the metabolic energy of the diet. The term NSP includes a wide group of compounds with different physicochemical properties, so it can be expected that the effects of these substances on poultry are also very different and wide. Nevertheless, in general, the anti-nutritional effects of these compounds are related to their sticky nature on the microbial population of the digestive system. These effects themselves cause changes in the passage time of nutrients from the intestine and also changes in hormonal settings due to a decrease in nutrient intake [10].

Therefore, according to the mentioned cases and the nutritional value of sorghum grain in poultry nutrition, as well as the use of edible enzymes in broiler rations to reduce anti-nutritional substances in feed and improve bird performance, this research aims to introduce sorghum grain as a useful feed in the diet of broiler chickens, as well as the use of enzyme as a useful and widely used additive in grain-based diet, the effect of these factors on the performance of broiler chickens can be investigated and studied scientifically.

Materials and Methods

This experiment was conducted in Qaimshahr city, Mazandaran province, Iran. Different levels of grain sorghum (0, 5, and 10%) and two levels of phytase enzyme and NSP (0 and 0.1%) were used in this study. April and May of 2021 for 42 days in a private broiler unit with a capacity of 40 thousand chickens, and at the end of the study, blood samples were taken and blood serum parameters were measured. During the test period, feed intake, daily weight gain, food conversion ratio, and carcass characteristics were measured. The experimental treatments include 1- control diet (without sorghum and no enzyme), 2- control diet + without sorghum + 0.1 enzyme, 3- control diet + 5% sorghum + without enzyme, 4- control diet + 5% sorghum + 1 0. enzyme, 5- control diet + 10% sorghum + no enzyme and 6- control diet + 10% sorghum + 0.1 enzyme. The statistical design used in this experiment was a 2x3 factorial method in the form of a completely random design, with 6 treatments and 3 repetitions (15 chickens in each repetition), and a total of 270 Ross 308 strain broiler chickens were used. Also, Excel and the SPSS software were used to analyze the data. The amount of feed consumed was determined, weighed, and provided to the birds in each cage. Chickens had free access to food and water during the experiment. The chicken breeding management program, including temperature, light, density, and bedding, was carried out by the recommended standard conditions. The vaccination program was carried out under the supervision of an experienced veterinarian.

The food rations of different experimental groups were adjusted based on the suggestions of the nutritional requirements tables of the Ross 308 strain using the UFFDA ration writing software based on corn and soybean meal. The experimental diets were: different levels of sorghum grain (zero, 5, and 10%) and three levels of phytase enzyme and NSP enzyme (zero and the amount recommended by the animal feed and poultry grain supplement factory) were used in feeding broilers.

Results and Discussion

Results of feed consumption: The amount of daily feed consumption is shown in Table 1. A statistically significant difference was observed in the main effect of sorghum in the first and sixth weeks and in the main effect of the enzyme only in the last week ($p<0.05$). In the first week, it was observed that the 10% sorghum treatment had the highest amount of feed consumed. Also, the lowest amount of feed consumed belonged to the 0% sorghum treatment. In the sixth week, it was observed that the treatment containing 0% sorghum had the highest amount of feed consumption and also the treatment containing 10% sorghum had the lowest feed consumption. The lower consumption of feed in treatments containing 5 and 10% sorghum compared to the control treatment (in the sixth week) is probably due to the presence of anti-nutritional factors such as tannin, which lowers palatability and makes the poultry less willing to eat.

In the main effect of the enzyme, feed consumption was higher in the treatment containing 0% enzyme and compared to the treatment containing 0.1% enzyme, the amount of feed consumption was lower (except for the sixth week). In the interaction effect of sorghum and enzyme, no statistically significant difference was observed between the treatments. Investigated the effect of sorghum along with enzyme in the diet on the performance of broiler chickens; the results showed that the replacement of sorghum along with enzyme did not have an adverse effect on feed consumption [11]. The results of the research of [12] showed that the addition of enzymes to the ration reduced feed consumption in the entire rearing period. Research on the effects of the late addition of phytase enzyme on broiler chickens showed that the addition of phytase enzyme significantly increased feed consumption ($p<0.05$) [13]. The results of Goli et al.'s research on the evaluation of the effects of enzyme supplements on the performance and blood serum metabolites of broiler chickens showed that in the growing and finishing periods, the feed consumption in the control treatment was reduced compared to the other groups [14].

Table 1. Feed intake measurement in different stages(g)

Treatments/weeks	Week-1	Week-2	Week-3	Week-4	Week-5	Week-6
The main effect of sorghum						
0	109.29^{b}	321.60	722.59	1256.05	2056.69	3489.14^{a}
5	122.02^{a}	341.81	712.97	1267.04	2024.19	33366.38^{b}
10	122.32^{a}	321.08	717.97	1245.65	2016.81	3296.20^{b}
The main effect of enzyme						
0	112.96	323.56	716.56	1245.82	2036.45	3435.05^{a}
0.1	117.78	332.77	718.54	1257.68	2042.01	3312.75^{b}
Mutual effects						
0 and 0	112.06	315.02	713.18	1254.6	2047.10	3680.27
0 and 0.1	106.52	328.18	731.99	1257.49	2066.28	3297.99
0 and 5	122.19	347.77	726.90	1271.22	2051.08	3340.95
0.1 and 5	121.84	335.84	699.040	1292.86	2037.28	3331.79
0 and 10	111.64	307.87	709.59	1238.62	2011.15	3283.93
0.1 and 10	124.99	334.28	724.57	1252.68	2022.46	3308.46
SEM	1.27	5.15	5.795	7.98	9.11	27.35
P. Value	0.009	0.31	0.59	0.89	0.59	0.009

Notes: [a,b] In each line, means with different superscripts are significantly different ($p<0.05$).

Weekly weight gains: The live weight gain results are shown in Table 2. In the main effect of sorghum, a statistically significant difference was observed only in the fifth and sixth weeks ($p<0.05$), so in weeks 2, 4, 5, and 6, the diet without sorghum had the highest amount of live weight. Sorghum has high tannin, making it difficult for digestive proteases to reach. High tannin reduces feed consumption. Decreasing consumption probably causes poor digestion of protein, which ultimately leads to poor weight gain. No statistically significant difference was observed in the interaction effect between sorghum and enzyme. Poultry does not produce enzymes for hydrolysis, and they remain unhydrolyzed [15].

These results reduce feed efficiency. The research of Antoniou [16] and Feighner [17] have shown that NSPs in the diet of broiler chickens negatively affect the microbial population of the digestive system, and this leads to a decrease in the digestibility and absorption of substances. The results of the research by Kriseldi [18] on the effect of correcting nutrients equivalent to phytase enzyme on the performance of broiler chickens showed that no significant effect was observed on body weight gain ($p>0.05$). Evaluated the effects of rapeseed meal degraded by enzymolysis and fermentation on broiler chickens and stated that these affect the weight gains of broilers significantly ($p<0.05$) [19].

The results of Ndazigaruye [20] on the evaluation of the effects of enzyme supplements on the performance and blood serum metabolites of broiler chickens showed that live weight gain in the initial period, growth, and final period and the entire breeding period in the control treatment showed a significant decrease compared to other groups containing enzyme ($p<0.05$).

Food conversion ratio: The results of food conversion ratio are shown in Table 3. Only in the main effect of sorghum, a statistically significant difference was observed in the fifth week ($p<0.05$). In the fifth week, the diet without sorghum had the highest food conversion ratio. In other weeks, diets containing 5 and 10% sorghum had a higher food conversion ratio than the control, and the high food conversion ratio in these treatments is probably due to the high levels of anti-nutritional factors in sorghum grains. Sorghum contains kafirin, phytate and tannin; these factors can negatively influence the nutritive. High tannin also reduces feed consumption. Decreasing consumption probably causes poor digestion of protein, which ultimately leads to poor weight gain [21].

Table 2. Live weight measurement in different stages(g)

Treatments/weeks	Week-1	Week-2	Week-3	Week-4	Week-5	Week-6
The main effect of sorghum						
0	88.61	191.45	395.29	754.25	1238.00^{a}	1755.50^{a}
5	89.61	188.59	397.88	753.22	1147.22^{b}	1607.77^{b}
10	86.96	180.51	372.92	720.00	1215.33^{ab}	1698.89^{ab}
The main effect of enzyme						
0	88.82	184.77	380.65	738.44	1226.37	1670.48
0.1	88.07	188.92	369.74	736.53	1174.00	1694.20
Mutual effects						
0 and 0	88.66	188.97	389.08	760.00	1179.77	1765.99
0 and 0.1	88.88	193.93	401.48	748.49	1114.66	1711.71
0 and 5	89.44	187.90	392.88	749.77	1209.66	1589.99
0.1 and 5	89.77	189.28	402.86	756.66	1221.00	1625.54
0 and 10	88.37	177.46	359.97	705.55	1289.66	1655.44
0.1 and 10	85.53	183.57	385.86	734.44	1186.33	1733.33
SEM	1.501	2.69	6.03	9.508	15.74	22.30
P. Value	0.96	0.61	0.40	0.60	0.12	0.25

Notes: ^{a,b} In each line, means with different superscripts are significantly different ($p<0.05$).

There was no statistically significant difference in the main effect of the enzyme and also the interaction between sorghum and the enzyme ($p>0.05$). Adamu et al showed that increasing the levels of sorghum in the diet improved the feed conversion ratio in broilers [22]. Also, Daramola showed that the mutual effects of sorghum and phytase enzymes had no significant effect on the feed conversion ratio of broiler chickens [11]. The research results of [12] showed that the addition of enzymes to the diet reduced the food conversion coefficient in the entire rearing period. The results of showed that enzyme addition had no significant effect on the feed conversion ratio (p>0.05) [13].

The results of the research of Kriseldi [18] on the effect of correcting nutrients equivalent to phytase enzyme on the performance of broiler chickens showed that no significant effect was observed on the feed conversion ratio in the growth and final period (p>0.05). The results of the research of Li et al, evaluated the effects of rapeseed meal degraded by enzymolysis and fermentation on broiler chickens and stated these affect the food conversion coefficient of broilers significantly (p 0.05) [19].

Table 3. Feed conversion ratio (FCR) measurement in different stages(g)

Treatments/weeks	Week-1	Week-2	Week-3	Week-4	Week-5	Week-6
The main effect of sorghum						
0	1.28	1.69	1.82	1.66	1.79a	1.92
5	1.36	1.81	1.85	1.68	1.68b	2.02
10	1.33	1.82	1.92	1.73	1.65b	1.94
The main effect of enzyme						
0	1.328	1.79	1.88	1.70	1.67	1.99
0.1	1.327	1.76	1.85	1.68	1.74	1.96
Mutual effects						
0 and 0	1.26	1.703	1.83	1.65	1.73	1.91
0 and 0.1	1.31	1.69	1.82	1.68	1.85	1.92
0 and 5	1.36	1.84	1.85	1.69	1.69	1.2
0.1 and 5	1.35	1.77	1.85	1.66	1.66	2.05
0 and 10	1.35	1.82	1.97	1.75	1.60	1.97
0.1 and 10	1.31	1.82	1.87	1.70	1.70	1.90
SEM	0.028	0.003	0.03	0.022	0.018	0.029
P. Value	0.89	0.43	0.88	0.80	0.04	0.34

Notes: ^{a,b} In each line, means with different superscripts are significantly different (p<0.05).

Weight of carcass components and internal organs: The results of the analysis of carcass components are shown in Table 4 and Table 5. Only in the main effect of sorghum in the liver, spleen, pancreas, small intestine, large intestine, and cecum, there was a statistically significant difference (p<0.05). In the main effect of the enzyme, carcass weight was higher in the treatment containing enzyme and it may be due to the increase in energy distribution to digestive organs (*ie*, gizzards and large intestine) and other internal organs (*ie*, liver and heart), which will also increase the heat and the total maintenance cost in broilers fed with these diets. These organs are relatively active, and their growth leads to an increase in heat production and, as a result, a decrease in the energy required to produce carcass weight. The highest fat in the abdominal area, liver, heart, spleen, pancreas, small intestine, and cecum was observed in the diet without sorghum. And the highest weight of carcass, wing, thigh, breast, back piece, gizzard, and large intestine were observed in the ration containing 10% sorghum, no statistically significant difference was observed in the rations containing enzyme (p>0.05). Adamu et al showed that as a result of substituting sorghum instead of corn in the diet of broilers, there was a statistically significant difference in live weight, pancreas, liver, cecum, and abdominal area fat among the treatments [22].

The results of effect of correcting nutrients equivalent to phytase enzyme on the performance of broiler chickens showed that a significant effect was observed on the percentage of the carcass and its components (percentage of breast, thigh and abdominal cavity fat) [18].

Table 4. Carcass traits measurement in different stages (g)

Treatments/weeks	Live weight	Thigh	Wings	Breast	Week-5	Abdominal fat
The main effect of sorghum						
0	1430.83	346	115.00	368	129.667	56.66
5	1369.50	282.67	118.83	369.67	120.66	50.167
10	1482.67	369.67	125.50	398.83	132.66	45.167
The main effect of enzyme						
0	1384.89	295.11	121.56	362	122.55	47.55
0.1	1482.44	370.44	118.00	395	132.77	53.77
Mutual effects						
0 and 0	1456.66	339.33	126.00	368.66	131.33	51.33
0 and 0.1	1405.00	352.66	104.00	367.33	128	62.00
0 and 5	1363.33	229.00	114.33	351.33	122.66	45.66
0.1 and 5	1375.66	336.33	123.33	338	118.66	44.66
0 and 10	1334.66	317.00	113.66	366	113.66	45.66
0.1 and 10	1630.66	422.33	137.33	431.66	151.66	54.66
SEM	33.75	18.99	5.904	14.68	3.437	3.437
P. Value	0.209	0.1868	0.669	0.684	0.66	0.66

Notes: ^{a,b} In each line, means with different superscripts are significantly different ($p<0.05$).

Table 5. Carcass traits measurement in different stages (g)

Treatments /weeks	Gizzard	Liver	Heart	Spleen	Pancreas	Small intestine	Large intestine	Cecum
The main effect of sorghum								
0	33.667	51.167^{a}	12.50	3.00^{a}	4.16^{a}	6.00^{a}	87.00^{b}	19.83^{a}
5	36.00	42.33^{b}	10.00	1.33^{b}	2.50b	3.33^{b}	92.167^{ab}	15.83^{b}
10	37.66	47.33^{ab}	11.00	2.00^{b}	2.83b	4.50^{ab}	105.50^{a}	15.00^{b}
The main effect of enzyme								
0	36.00	47.44	10.44	2.00	3.66	5.44	97.11	17.00
0.1	35.55	46.44	11.88	2.22	2.66	3.77	92.66	16.77
Mutual effects								
0 and 0	29.00	57.33	12.66	3.33	5.33	8.66	102.33	20.00
0 and 0.1	38.33	45.00	12.33	2.66	3.00	3.33	71.66	19.66
0 and 5	39.00	44.66	9.66	1.00	2.33	2.66	95.66	16.66
0.1 and 5	33.00	44.00	10.33	1.66	2.66	4.00	88.66	15.00
0 and 10	40.00	40.33	9.00	1.66	2.33	5.00	93.33	14.33
0.1 and 10	35.33	54.33	13.00	2.33	2.33	4.00	117.66	15.66
SEM	2.09	1.08	0.474	0.184	0.24	0.47	3.27	0.747
P.Value	0.65	0.002	0.127	0.03	0.03	0.03	0.03	0.201

Notes: ^{a,b} In each line, means with different superscripts are significantly different ($p<0.05$).

The biological criteria for the unbalanced distribution of nutrients to the different organs are not understood in the present study. Similarly, did not find any significant difference in the breast weight of ducks that were fed with liquor distiller grains [23]. The pH for chickens was in the normal range (5.5-6.5), which shows that the inclusion of malted sorghum in the diet of broilers does not affect the glycogen level [24]. The pH range reflects the amount of glycogen in the breast muscle before slaughter and how quickly the remaining glycogen is converted to lactic acid after slaughter [25]. The liver may be overworked to detoxify the chemicals in sorghum. In general, an increase in the size of the liver and heart can indicate the need to deal with toxic substances in the feed [26]. Larger stones can be explained by the higher structural wall components of malted sorghum powder (28.6% crude fiber) reported by Moses et al or natural particles in the diet [27].

Table 6. Blood traits measurement in different stages(g)

Blood samples/treatments	GLU	TG	CHLO	HDL	LDL	Ca	P	TP
The main effect of sorghum								
0	251.83	88.42	131.66^{a}	79.66^{a}	45.00^{a}	10.30	5.85	1.72
5	226.67	87.00	113.33^{b}	65.25^{ab}	36.41^{c}	9.45	5.30	1.52
10	214.33	66.58	124.83^{a}	73.66^{b}	40.417^{b}	9.75	6.167	1.63
The main effect of enzyme								
0	239.89	86.67	124.77	73.77	40.77	9.82	5.88	1.69
0.1	222.000	74.67	121.77	71.94	40.44	9.66	5.66	1.56
Mutual effects								
0 and 0	204.33	72.50	135.66	83.00	45.16	10.33	5.86	1.91
0 and 0.1	224.33	60.66	127.66	76.33	44.83	9.73	5.83	1.53
0 and 5	284.00	82.16	112.33	65.66	34.83	8.60	5.58	1.50
0.1 and 5	219.66	94.69	114.33	64.83	38.00	10.30	5.02	1.55
0 and 10	231.33	69.33	126.33	72.66	42.33	10.53	5.53	1.66
0.1 and 10	222.000	104.66	123.33	74.66	38.50	8.96	6.80	1.61
SEM	1.24	6.24	1.401	1.708	0.56	0.29	0.41	0.062
P.Value	0.46	0.37	0.038	0.075	0.001	0.35	0.88	0.47

Notes: [a,b] In each line, means with different superscripts are significantly different ($p<0.05$).

Blood parameters: The results of blood parameters analysis are shown in Table 6. Only in the main effect of sorghum, a statistically significant difference was observed in cholesterol, HDL, and LDL levels ($P<0.05$). The highest and lowest levels of cholesterol, HDL, and LDL were observed in diets without sorghum and diets containing 5% sorghum, respectively. No statistically significant differences were observed in diets containing enzymes and the mutual effects of sorghum and enzymes. Studied the effect of different levels of extruded soybeans and enzymes on broiler chickens, the results showed that blood parameters including blood cholesterol and triglycerides, liver and heart weight were affected by different grain levels [28]. The effect of correcting nutrients equivalent to phytase enzyme on the performance of broiler chickens showed that no significant effect was observed on the concentration of phosphorus in blood serum in experimental diets ($p>0.05$) [18].

Conclusion

The results of this research showed that the use of sorghum grain when increased 10% of the diet had no significant effect on live weight and feed conversion ratio, but it caused a decrease in feed consumption in the sixth week of the study. The effect of the enzyme could not cause a significant improvement in feed consumption, feed conversion ratio, and average live weight of chickens. As a result, it can be concluded that sorghum grain up to 10% level can be used in the diet of broiler chickens without any negative effects on the performance of broiler chickens.

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