



Effects of multienzymes in reduced energy diet on broiler chickens' performance under heat stress

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Received:	Abstract
Jan. 17, 2025	This study aims to investigate the multienzymes use , in broiler chicken under heat stress with low energy to estimate the Productive performance (body weight, feed intake, feed conversation ratio and body weight gain) ,The experiment was conducted from December 27, 2023 to February 1, 2024. In a private field the AlIbrahimiyyah area of Karbala, where the conditions were suitable for standard poultry farming, a total of 250 one-day-old broiler chickens of Rose 308 breed, with unsexed, were taken and divided into five groups. The first group consisted of 50 birds that Negative - control were (NCON) fed a basic diet without any additives and were kept away from heat stress, the second group consisted of 50 birds that were Positive control (PCON) fed a basic diet without any additives under heat stress .The third group consisted of 50 birds that were fed a basic diet with the addition of multienzymes under heat stress (MHS), the fourth group consisted of 50 birds that were fed a basic diet with the addition of multienzymes and reduced energy under heat stress) MLHS), the fifth group consisted of 50 birds that were fed a basic diet without additives under heat stress with reduced energy (LHS), After completing the experiment and collecting samples from chickens, a total of 20 samples from all replicates, the analysis results were as follows.
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Introduction

In intensive broiler production, birds are exposed to various stress factors, mainly heat stress [1]. Heat stress is one of the severe stress factors that negatively influence broiler productivity in tropical and subtropical regions, especially in humid environments [2]. At the same time, heat stress causes physiological abnormalities in the endocrine and immune systems and electrolyte imbalances, which are detrimental to the health and production of broilers [2]. Heat stress also damages gut structure and function (including low intestinal epithelium regeneration and integrity), decreasing broilers' growth performance and metabolic rate [3] Exogenous protease supplementation has been shown to improve the digestibility of crude protein and amino acids in poultry

diets, helping to reduce feed costs, environmental impact, and alterations to the chicken gut ecosystems. The supplementation of chicken diets with combinations of xylanases and proteases has been extensively investigated [4].

Nutrient digestibility and growth performance have been positively impacted by enzyme supplementation. However, the beneficial effects on the performance of diets based exclusively on wheat and soy seem to be less durable or dose-dependent. It has been shown that integrating maize-soybean diets with more diverse NSP-degrading enzyme activities improves broiler performance, although this approach may lack consistency. Dietary energy is one of the most expensive nutrients, along with amino acids, in chicken diets [5, 6, 7, 8].

Materials and Methods

This study was conducted indoors during the period from December 27, 2023 to January 31, 2024. A total of 250 one-day-old Ross 308 chicks (unsexed) were used, and they were divided into five groups, each group containing 50 birds with two replicates for each group. Each replicate consisted of 25 birds per cage, and the experimental treatments were as follows: 50 chicks control negative (NCON) fed corn-soybean diet only without HS, 50 chicks fed basal + multienzymes under HS, 50 chicks control positive (PCON) fed corn-soybean diet under HS, 50 chicks fed basal diet + multienzymes with reduced energy under HS, and 50 chicks fed with reduced energy under HS. The experiment lasted for five weeks, during which feed and water were freely provided to the birds during the study period.

Specific programs were followed to vaccinate the birds and take care of their health according to the recommendations for raising broiler chickens. A lighting system was adopted that included 23 hours of light and one hour of darkness, and the chicks were exposed to heat stress from the first day by increasing the temperature by 6°C above the normal rate, and this continued throughout the day until the end of the experiment using a gas-heated incubator.

Results and Discussion

The results of the current study, in the tables (4-1) and (4-2), showed a significant ($p \leq 0.05$) increase in the live body weight and weight gain in chickens. Starting in week one, from T1 to T2, there is a decrease in weight (183.62 to 176.52). The weight then increased at T3 and T4, indicating a positive response to multienzymes, as the weight increased to 190.06 and 188.14, respectively. At T5, there was a slight decline again to 181.88.

Means with a different small letter in the same column are significantly different ($P < 0.05$), means with a different capital letter in the same row are significantly different ($P < 0.05$). The second week, the live body weight group T3 had the highest weight (442.40 g), while T2 had the lowest (399.58 g), confirming the additional benefit of enzymes under heat stress. The third week again, the T3 group had the highest weight

(1025.92 g), while the T2 group had the lowest (822.26 g). The difference was significant between the groups with enzymes and the group exposed to heat stress without enzymes. In the fourth week, the difference in live body weight became more noticeable, with the T3 group recording the highest weight (1561.42 g), while T2 and T1 were much lower (1306.78 and 1425.50 g).

The fifth week in the end, T3 recorded the highest weight (2189.12 g), followed by T4 (2071.48 g). T2 and T5 had the lowest weight (1920.0, 1924.14 g), which indicates that low energy or not adding enzymes negatively affects weight under heat stress.

Table (2): Effect of multienzymes on broiler chickens live body weight (gm).

/Group weeks	T1	T2	T3	T4	T5
Day1	43.74±0.30 A	43.76±0.51 A	43.40±0.64 A	43.20±0.42 A	43.08±0.54 A
Week1	183.62±4.23 B	176.52±3.59 C	190.06±5.36 A	188.14±2.40 A	181.88±1.92 B
Week2	417.54±3.54 B	399.58±6.02 C	442.40±12.23 A	435.68±8.61 A	424.20±6.74 B
Week3	909.38±14.60 C	822.26±8.28 D	1025.92 ±5.72 A	976.46±8.04 B	905.84±2.57 C
Week4	1425.50 ±10.5 C	1306.78±5.94 E	1561.4±15.53 A	1524.44±7.49 B	1413.64±6.61 D
Week5	1948.38 ± 4.25 C	1920.0 ± 24.49 D	2189.12 ±7.91 A	2071.48±31.26 B	1924.14±3.58 D

Different letters among groups showed a significant difference at ($p \leq 0.05$). Negative - control (NCON) (T1) fed on a basil diet without HS (T2) Positive control (PCON) fed on a basil diet subjected under H, (T3) fed basil diet with multienzymes under HS (MHS), (T4) fed basil diet with multienzymes subjected reduced energy under HS (MLHS), (T5) fed basil diet with low energy subjected under HS (LHS).

Table (3): Effect of multienzymes on broiler chickens weight gain (gm) (Mean ± SE).

/group weeks	T1	T2	T3	T4	T5
Week1	139.87±4.02 B	132.76±3.39 C	146.66±5.09 A	144.94±2.44 A	138.80±1.72 B
Week2	233.92±6.95 BC	223.06±8.44 C	252.34±16.8 4 A	247.54±7.63 A	242.32±6.0 AB
Week3	491.84±16.9 6 C	422.68±9.90 D	583.52±13.5 8 A	540.78±15.3 0 B	481.64±6.9 C
Week4	516.12±16.0 3 B	484.52±7.26 C	535.50±12.7 57 A	547.98±10.4 2 A	507.80±5.2 B

Week5	522.88±8.88 C	613.22±19.97 A	627.70±18.4 5 A	547.04±33.1 1 B	510.50±6.0 C
WG cum	1904.63±4.1 C	1876.24±24.05 D	2145.72±7.8 9 A	2028.28±31. 4 A	1881.06±3.3 D

Different letters among groups showed a significant difference at ($p \leq 0.05$). Negative - control (NCON) (T1) fed on a basil diet without HS (T2) Positive control (PCON) fed on a basil diet subjected under H, (T3) fed basil diet with multienzymes under HS (MHS), (T4) fed basil diet with multienzymes subjected reduced energy under HS (MLHS), (T5) fed basil diet with low energy subjected under HS (LHS).

The results in (table 4) indicated that in the first week, the highest feed consumption was in T1 (177.08 g) and T3 and T4 (178.24, 178.40 g), which means that chicks that were not exposed to heat stress or that took enzyme supplements had significantly higher feed consumption; T2 (169.96 g) was the lowest in feed consumption, which reflects the negative effect of heat stress. The second week: T3 continued to record the highest feed consumption (353.02 g), followed by T1 (349.12 g), while T2 had the lowest feed consumption (338.16 g), and T4 recorded moderate feed intake (250.48 g), which may reflect energy efficiency with nutritional supplementation.

The third week: T1 and T3 again recorded the highest feed consumption (803.50 g and 799.32 g respectively), indicating that multiple enzymes help maintain feed consumption similar to normal feeding. T2 was clearly the lowest (723.10 g), indicating a negative effect of heat stress on birds' appetite. The fourth week, T1 continued to record the highest feed consumption (987.04 g), while feed consumption decreased significantly in T2 (838.06 g). T3, although under heat stress, maintained good feed consumption compared to the rest of the groups (851.62 g). The fifth week feed consumption in all groups was almost equal, with T1 and T3 groups recording very similar intake (994.38 g and 989.80 g, respectively), and T4 recorded the lowest feed consumption this week (978.36 g).

The results in table (5) for the first week: group T3 (1.21) was the best in terms of feed conversion efficiency, indicating that multiple enzymes under heat stress improved conversion efficiency; T2 (1.28) recorded the highest conversion rate, which means that thermal stress negatively affects efficiency. Other groups had moderate results. The second week: Group T3 (1.40) and Group T4 (1.41) recorded the best results this week, reinforcing the role of multiple enzymes in improving efficiency under stress conditions. T1 (1.49) and T2 (1.51) were less efficient in converting feed compared to groups containing enzymes.

The third week: T3 recorded the best conversion rate (1.37) followed by T4 (1.43), indicating that multiple enzymes help maintain excellent conversion efficiency even with heat stress. T2 recorded the highest conversion rate (1.71), reflecting the effect of negative thermal stress. The fourth week: group T3 (1.59) was the best this week as

well, with a noticeable improvement in conversion efficiency. T2 and T1 had the highest conversion rates (both 1.73), indicating that heat stress strongly affects conversion efficiency in the absence of enzyme supplementation.

The fifth week: T3 (1.57) and T4 (1.79) had good conversion rates compared to the rest of the groups. T1 and T5 had the highest conversion rates (1.90 and 1.93, respectively), indicating lower feed conversion efficiency.

Table (4): Effect of multienzymes on broiler chickens Feed intake (gm) (Mean \pm SE)

Group\ weeks	T1	T2	T3	T4	T5
Week 1	177.08 \pm 1.20 A	169.96 \pm 2.25 C	178.24 \pm 3.12 A	178.40 \pm 1.93 A	173.86 \pm 1.29 B
Week 2	349.12 \pm 1.26 B	338.16 \pm 1.52 D	353.02 \pm 2.27 A	250.48 \pm 0.99 B	344.92 \pm 0.68 C
Week 3	803.50 \pm 1.70 A	723.10 \pm 3.42 D	799.32 \pm 6.42 A	773.54 \pm 8.90 B	749.86 \pm 4.25 C
Week 4	987.04 \pm 1.97 A	838.06 \pm 4.33 C	851.62 \pm 2.57 B	848.56 \pm 4.16 B	848.44 \pm 2.68 B
Week 5	994.38 \pm 1.73 A	989.60 \pm 9.87 A	989.80 \pm 3.66 A	978.36 \pm 3.84 B	988.22 \pm 4.56 A
FI cumulative	3221.12 \pm 2.3 A	3035.86 \pm 5.13 E	3172.00 \pm 7.55 B	3129.34 \pm 10.11 C	3105.30 \pm 6.3 D

Table (5): Effect of multienzymes on Feed conversion ratio in broiler chickens (Mean \pm SE)

Group\ weeks	T1	T2	T3	T4	T5
Week 1	1.26 \pm 0.04 AB	1.28 \pm 0.03 A	1.21 \pm 0.06 C	1.23 \pm 0.009 C	1.25 \pm 0.02 C
Week 2	1.49 \pm 0.04 A	1.51 \pm 0.05 A	1.40 \pm 0.82 B	1.41 \pm 0.04 B	1.42 \pm 0.03 B
Week 3	1.63 \pm 0.05 B	1.71 \pm 0.03 A	1.37 \pm 0.03 E	1.43 \pm 0.04 D	1.55 \pm 0.02 C
Week 4	1.73 \pm 0.05 A	1.73 \pm 0.02 A	1.59 \pm 0.03 C	1.54 \pm 0.02 D	1.67 \pm 0.02 B
Week 5	1.90 \pm 0.03 A	1.61 \pm 0.04 C	1.57 \pm 0.04 C	1.79 \pm 0.11 B	1.93 \pm 0.02 A
mean	1.69 \pm 0.004 A	1.61 \pm 0.01 C	1.47 \pm 0.006 E	1.54 \pm 0.02 D	1.65 \pm 0.003 B

Different letters among groups showed a significant difference at ($p \leq 0.05$). Negative - control (NCON) (T1) fed on a basil diet without HS (T2) Positive control (PCON) fed on a basil diet subjected under H, (T3) fed basil diet with multienzymes under HS (MHS), (T4) fed basil diet with multienzymes subjected reduced energy under HS (MLHS), (T5) fed basil diet with low energy subjected under HS (LHS).

The study showed that the effect of the addition of xylanase to diets containing dried corn grains with solubles (sDDGS) significantly reduced the concentration of insoluble NAP and increased the concentration of free sugars (arabinose and xylose) in the ileal digesta, and the availability of these free sugars may have provided nutrients to the bird, thus improving the feed conversion rate. In the present study, the addition of xylanase to a wheat-based diet significantly increased the ileal and total digestibility of soluble and insoluble NSPs this agree with [10].

Therefore, it can be concluded that exogenous xylanase eliminates the antinutritional effect of NSP through partial hydrolysis of soluble and insoluble NSP, resulting in a decrease in the viscosity of the ileal digesta and an increase in the digestibility of nutrients. In addition, the higher total digestibility of soluble and insoluble NSPs compared with those in the ileal digesta may be due to caecal fermentation [11]. It is well known that water-soluble arabinoxylan is one of the main antinutritional agents of PNA in wheat-based diets, which increases the viscosity of digestate, preserves nutrients, alters the intestinal microflora, interfering with digestion and absorption of nutrients. [12]

Which may be the direct cause of improved growth performance in birds. Our results are partially consistent with the results of [13] who reported that xylanase significantly reduced digestate viscosity and improved apparent macronutrient digestibility of dry matter, milk protein and energy in broilers fed grain diets from 1 to 35 days [3].

The present results, like those of [14], suggested that xylanase can reduce the elimination of nutrients and increase their digestibility, mainly starch and proteins instead of fats. Increase intestinal viscosity, but it also changes the functions of the digestive tract by modifying the secretion of internal digestive enzymes, water and electrolytes, and increasing fermentation in the small intestine [15]. Soluble and insoluble NSP, which leads to a decrease in the viscosity of intestinal digestion and an increase in the digestibility of nutrients. In addition, the digestibility of soluble and insoluble NSPs in the small intestine may be higher than in ileal digestion, mainly due to cecal fermentation [16].

Increased intestinal viscosity and the prevalence of pathogenic bacteria in the small intestine, causing greater inflammation and oxidative stress in birds. Diets containing wheat, barley and rye can be improved with multienzymes, suggesting that the adverse effects of these grains may be partly attributable to an increase of intestinal viscosity will lead to proliferation of pathogenic bacteria of small intestine It has been suggested that the use of carbohydrate enzymes to compensate for the negative effects of NSPs may increase intestinal viscosity, leading to increased pathogenic bacteria, which reduces the incidence of these pathogenic bacteria that lead to the negative impact on

feed intake [17]. The use of carbohydrase enzymes is used to compensate for the negative effects NSPs have been used since the late 1980s to report ant nutritional effects. Arabinoxylans and β -glucanases are used to reverse the ant nutritional effects of β -glucans. The reduction in viscosity leads to improved poultry performance and nutrient digestibility [18]. The reduction of digestate viscosity by enzyme supplementation plays a more important role in young birds than in their older counterparts. This is thought to be because young birds have a more immature intestinal tract and it is assumed that young birds produce less pancreatic enzymes. Non-starch polysaccharides can have a negative effect on poultry health due to an increased risk of infection from competition pathogenic bacteria present in the intestinal microbiota for digestible nutrients.

Non-starch polysaccharides have been reported to increase solvent viscosity in the small intestine, leading to a decrease in the absorption and digestibility of nutrients. This slippery chemistry has led to an overgrowth of pathogenic bacteria, intestinal inflammation, and altered intestinal barrier function, which can result in increased oxidative stress and inflammation. It has been repeatedly reported that the inclusion of xylanase, especially in diets rich in wheat, reduces the viscosity of jejunal and ileal digesta. In addition, xylanases increase the rate of digestion and the digestion of nutrients in the small intestine, which limits the growth of fermented microorganisms [16,19].

β -Glucanase is a type of cellular enzyme commonly produced by bacteria, fungi, and various actinomycetes. β -Glucanase is not naturally produced by non-ruminants, but can be found in rumen bacteria of ruminants. β -Glucanase hydrolyzes the glycosidic bonds of β -glucans and reduces the antinutritional effects of β -glucans [16]. Amylase is an enzyme that digests starch to make more energy available by increasing starch digestion. Reported that during the first 7-day period, growth performance of birds fed amylase improved by 9.4% and feed conversion by 4.2[20].

Suggest that in the first week, after hatching, the chicks have a higher sensitivity to amylase because they need the help of the enzyme to compensate for their immature production of pancreatic amylase due to the development of the most important part of his intestinal tract. Evidence supports that, compared to their juvenile counterparts, older birds produce more pancreatic amylase [21]. However, this does not mean that older birds do not benefit from amylase supplementation. Amylase supplementation has also been shown to improve dietary AMEn [22,23]. reported that α -amylase decreased jejunal viscosity while increasing the total digestibility of starch and energy in the gastrointestinal tract. Similarly, amylase has improved the use of energy when added to the diet of corn and soy flour. This is also suggested by the evidence increasing amylase levels linearly increases the ileal digestibility of resistant starch [21]. The results showed that xylanase enzymes have a significant effect WG and FCR ($p < 0.05$) in the growing and finishing stages, as well as in the whole breeding season. These results agree with those of [24,25]. It has also been reported that exogenous xylanase improved nutrient digestibility and growth performance in maize-based poultry diets

[26]. It seems that xylanase breaks the cell wall and liberates more nutrients for digestion and absorption.

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