

Effects of Dietary *Moringa oleifera* Oil on Ammonia-Induced Physiological Stress in Broiler Chickens.

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1. Abstract

Inadequate ventilation in poultry farms leads to increased ammonia levels which can endanger the health of broiler chickens through oxidative damage, respiratory diseases, and metabolic disorders. This study evaluated the efficacy of dietary inclusion of *Moringa oleifera* oil (MO) as a means of alleviating physiological stress on broilers through high ammonia exposure. A total of 180 one-day-old male Ross 308 broilers were used in a 2 x 3 factorial design with 2 feed treatments (control diet versus diet supplemented with 10 g/kg MO) compared across 3 environmental treatments (normal air, moderate ammonia, and severe ammonia). The environmental stressor was applied from 14 to 21 days of age). Ammonia exposure adversely influences physiological performance and organ function in regards to the right ventricular hypertrophy and lung hypertrophy, serum liver enzyme concentrations (increased AST; ALT), cellular damage indices (increased LDH), and lipid profiles of broiler chickens. However, 1.0% *Moringa oleifera* (MO) supplementation mitigated the adverse effects of ammonia by producing significantly reduced serum AST, ALT, and LDH levels, and significantly decreased serum creatinine and uric acid levels, and by producing improved serum lipid profiles, as indicated by significantly lower serum concentrations of LDL and triglycerides, compared to ammonia-exposed (non-MO) birds. *Moringa oleifera* supplementation reduced hepatic congestion and intestinal inflammation. Collectively, these results suggest that the antioxidant properties of *Moringa oleifera* support broilers in maintaining organ function and metabolism during ammonia environmental stress.

Keyword: Ammonia stress, Broiler chickens, *Moringa oleifera* oil, Oxidative stress, Serum biochemical parameters

2- Introduction

Poultry farming has become more intensive, and consequently the production of poultry meat as a foundation for food security is accomplished with an increased number of birds stocked in high-density indoor housing systems according to the demand. These

high-density production systems, however, severely limit the amount of environmental enrichment available to birds and, as a result, lack sufficient ventilation. Insufficient ventilation results in birds being exposed to high levels of noxious gases, primarily ammonia (NH₃) due to the breakdown of uric acid by microorganisms.

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High atmospheric ammonia is an ongoing challenge in modern broiler houses, posing serious health and productivity threats to the birds (1; 2). Furthermore, modern broiler strains are genetically selected for fast growth, resulting in rapid metabolic rates that make them highly susceptible to ammonia toxicity. Therefore, strict environmental control is critical for maintaining production efficiency in these intensive farming systems

It is widely accepted that exposure to elevated levels of ammonia over long time periods or multiple times can be extremely harmful to the respiratory system of broilers. High levels of ammonia can damage the respiratory epithelial lining, disrupt the mucociliary clearance of mucus from the airways to the outside of the body, and can also increase a bird's likelihood of developing respiratory disease, as well as disrupt the way they use oxygen and perform physiologically (3; 4). Long-term exposure to high levels of ammonia causes activation of inflammatory processes in both the lungs and the air sacs, which can subsequently adversely affect how much effort is used to work the heart and can increase metabolic stress on birds which will likely show up as decreased performance (5; 6).

High exposure levels to ammonia will also bring on systemic stresses, which not only creates respiratory damage but can trigger impact on multiple organ systems and metabolic pathways. Elevated levels of ammonia within the blood set off oxidative stress and a systemic inflammatory response, ultimately leading to damage to cells and tissues throughout the body. In general, inflammatory responses that occur as a result of ammonia exposure can be

assessed based on alterations in blood hematological profile such as increased serum levels of alanine aminotransferase (ALT) and aspartate aminotransferase (AST) and alteration in serum levels of lactate dehydrogenase (LDH) indicate strain on the liver and generalized cell damage (7; 8). Ammonia stress alters lipid metabolism and thus affects triglycerides, total cholesterol, and lipoprotein fractions such as HDL, LDL, etc. There will be relative organ weight discrepancies affecting organs such as the liver, heart and lungs as these organs have altered to meet the metabolic demands placed upon them by the beneficial and deleterious effects of exposure to ammonia (2; 9).

The inclusion of moringa leaves, seeds, and/or oil into broiler diets at normal production rates has been found to positively impact growth performance, feed efficiency, and oxidative metabolism (10; 11; 12; 13). Moreover, the supplementation of moringa has also been shown to increase the activity of antioxidant enzymes, modulate immunity, and reduce markers of oxidative damage (14; 15).

Despite the fact that ammonia has negative effects on the physiology of broilers, and that there is growing evidence to suggest that moringa provides benefits, there is very little direct information regarding the protective capacity of moringa against ammonia-induced damage to the physiology of broilers. Research has established that multiple antioxidants (including vitamin E, selenium, L-carnitine, and numerous plant extracts) have successfully reduced oxidative injuries caused by ammonia stress and have reduced inflammation in the animals (16; 17; 18). Therefore, the specific effectiveness of moringa oil for the protection of broilers against ammonia

toxicity due to inadequate ventilation has yet to be sufficiently explored. Moringa has been suggested as a potential supplement for dietary oil to improve physiological disruption due to ammonia exposure and aid in the maintenance of organ function due to its high levels of anti-inflammatory compounds and antioxidant activity. This

3. Materials and Methods

3.1 Birds, Dietary Treatments, and Husbandry

The experiment was conducted at the Poultry farm of Department of Animal Science, College of Agricultural Engineering Sciences, University of Sulaimani. The experiment was conducted between 10th march to 20th April.

A commercial hatchery provided a total of 180 one-day-old male Ross 308 broiler chicks to the livestock facility. Upon arrival at the livestock facility, the chicks were weighed and randomly assigned to one of six different treatment groups. The formulation of both experimental diets included a basal diet that met or exceeded the [19] minimum requirement for calorie

- **CON:** Control basal diet (containing standard soybean oil).
- **MO:** Moringa oil-supplemented diet (10 g/kg).

The birds had free access at all times throughout the 35-day trial period to mash feeds as well as fresh drinking water. Throughout the 35-day trial the photoperiod was 23 hrs. continuous light and 1 hr. darkness.

3.2. Experimental Design and Induction of High Ammonia Stress

The study was a 2 × 3 factorial study design consisting of two different dietary

study aimed to investigate the protective effects of dietary *Moringa oleifera* oil supplementation against ammonia-

induced physiological stress in broiler chickens by evaluating organ indices, liver and kidney function, and serum lipid metabolism.

(Nutritional Energy) and lipid (Fat). Both diets contained corn and soybean as their primary source of calories (Nutritional Energy). Cold-Pressed Moringa Oil at a level of 10 g/kg (1.0%) was also included in the basal diet as part of both experimental diets. Moringa Oil was incorporated into the basal diet to replace an equal amount of soybean oil; both experimental diets were formulated to provide iso-caloric (calories) and iso-lipidic (fat) diets. The liquid oils were blended with the feed using a horizontal mixer to ensure complete product homogeneity. The two experimental diets are as follows:

treatments (Control vs. MO) under three different environmental conditions (Normoxic, Moderate Ammonia and Severe Ammonia). This provided a total of six experimental groups:

1. **Normoxia + CON:** Control diet under normal ventilation.
2. **Normoxia + MO:** Moringa oil diet under normal ventilation.

3. **Moderate Ammonia + CON:** Control diet under moderate Ammonia stress.
4. **Moderate Ammonia + MO:** Moringa oil diet under moderate Ammonia stress.
5. **Severe Ammonia + CON:** Control diet under severe Ammonia stress.
6. **Severe Ammonia + MO:** Moringa oil diet under severe Ammonia stress

Table 1: Ingredient composition of the control and Moringa oil-supplemented diets fed to broiler chickens (g/kg).

Ingredients	Starter	Grower	Finisher
Soya bean meal	360	308	252
Yellow Corn	500	450	450
Premix*	25	25	25
Soybean Oil	10	15	17
Moringa Oil	10	10	10
Limestone	15	12	10
Wheat	77	177	233
Salt	1	1	1
Toxin binder	2	2	2
Chemical Analysis			
Protein (%)	22.6	20.2	18.01
Metabolizable Energy (kcal/kg)	3000	3100	3150
Ash	7.26	5.17	4.34
Fat	4	4.84	5.15
Fiber	2.68	2.68	2.58
Moisture	8.88	10.75	11.32

*dofamix Premix consist of :17% crude protein,0.6% crude fiber, 0.37%crud fat, 70% crude ash, 14.4calcium, 3.65phosphorus, 7%lysine, 9%methionine, 1%threonine. Trace elements: 2,400 mg Fe (3b103 Iron (II)sulphate monohydrate); 80 mg I (3b202 Calcium iodate); 600 mg Cu (E4 Cupric (II)sulphate-pentahydrate); 2,800 mg Mn (3b502 Manganese (II)oxide); 2,800 mg Zn (3b605 Zinc sulphate monohydrate); 10 mg Se (E8 Sodium Selenite).

The 180 birds used in this study were divided into six groups with three replicates of 10 birds. An environmental challenge was given during the period between day 14 and day 21 after hatch.

The environmental conditions were established as follows:

- **Normal Ammonia (<10 ppm):** Birds were kept in identical room similar to other groups but with zero added ammonia.
- **Moderate Ammonia (25 ppm):** Birds were housed in custom-built room.

- **Severe Ammonia (50 ppm):** Birds were housed in fully controlled environmental room.

we have used an **all-in-one industrial dual-relay controller** for each room. To build one independent room, I needed to source these four interconnected components:

- 1- **The Controller:** A wall-mounted gas monitor with an internal sensor and **two independent mechanical relays** (an *Evikon PluraSens E2611-NH3*).
- 2- **The Dosing Actuator:** A Valve (**Bürkert Type 6013**) rated for ammonia service. It connected to the tank of ammonia in the room and it opens when powered to introduce gas and snaps shut instantly if power fails.
- 3- **The Exhaust Actuator:** A fan to vent excess gas, the fan of the room was connected to one of the two built in relay of the wall mounted gas monitor to turn on and off the fan.
- 4- **Intermediate Power Supplies:** Standard industrial **110V/220V AC to 24V DC Power Supply** (Mean Well HDR-15-24 power adapters) to separate the control signals from the heavy driving power. This is needed because the ammonia meter runs on its own power source but the gas controller's built-in relay does not generate electricity; it only acts as a switch.

Setting Up the Chambers

Once we wired the four components of **all-in-one industrial dual-relay controller**, I used the controller's digital interface menu (*Evikon PluraSens E2611-NH3*) to program my specific parameters per room:

- **Room 1 Setup (the birds were raised under normal air condition and this room didn't need any setup)**
- **Room 2 Setup (Target: 25 ppm)**
 - *Relay 1 (Dosing) Trigger:* Set to turn ON at **23 ppm** and OFF at **25 ppm**.
 - *Relay 2 (Exhaust) Trigger:* Set to turn ON at **27 ppm** and OFF at **25 ppm**.
- **Room 3 Setup (Target: 50 ppm)**
 - *Relay 1 (Dosing) Trigger:* Set to turn ON at **48 ppm** and OFF at **50 ppm**.
 - *Relay 2 (Exhaust) Trigger:* Set to turn ON at **52 ppm** and OFF at **50 ppm**.

The control system-maintained ammonia concentrations within the predetermined ranges for each treatment room, independent of ammonia originating from litter decomposition.

The temperature ($25 \pm 1^{\circ}\text{C}$) and relative humidity ($60 \pm 5\%$) were kept constant across all chambers and pens during the challenge period. Ammonia, oxygen, and carbon dioxide concentrations were monitored twice daily (08:00 and 16:00 h) using a calibrated handheld multi-gas analyzer (**GASLABPLUS CM-500 series, Henan Zhongan Electronic Detection Technology Co., Ltd, Zhengzhou, China**). Feed and water were available *ad libitum*.

3.3. Data Collection and Sample Processing

3.3.1. Sample Collection and Organ Indices

After completing the stress period (22nd day), individual birds were put down with a painless method by dislocating their spinal cord in the cervical region (neck area of the bird). Blood samples (5 mL) were taken using serum separator tubes. After necropsy, organs (and intestines) were removed and weighed for use in calculating relative organ weights as a % of live body weight.

3.3.2. Serum Biochemical Analysis included

- **Liver Function:** Alanine Aminotransferase (ALT) and Aspartate Aminotransferase (AST).
- **Kidney Function:** Creatinine and Urea.
- **General Cellular Damage:** Lactate Dehydrogenase (LDH).
- **Lipid Metabolism:** Triglycerides, Total Cholesterol, and High-Density Lipoprotein (HDL). Low-Density Lipoprotein (LDL) was calculated using the Friedewald equation.

3.4. Statistical Analysis

Data were analyzed as a 2×3 factorial arrangements using the General Linear Model (GLM) procedure of XLStat (v2019.2.2). The statistical model was:

4. Results and Discussion

4.1 Effects of Ammonia Exposure and Moringa Oil on Organ Indices

There was no observed impact of *Moringa oleifera* oil supplementation on relative organ weight in broiler chickens (table two); however, ammonia concentration of air

$$Y_{ijk} = \mu + \alpha_i + \beta_j + (\alpha\beta)_{ij} + \varepsilon_{ijk}$$

Where:

- Y_{ijk} = the observed value for a specific variable
- μ = the overall mean
- α_i = the main effect of diet (CON vs. MO)
- β_j = the main effect of environment (Normoxic, Moderate, Severe)
- $(\alpha\beta)_{ij}$ = the interaction effect between diet and environment
- ε_{ijk} = the random experimental error

The study was done using replicate pens. The impact of treatments was analyzed, as well as any interaction effects, and established the significance of these impacts (main effects) using a statistical analysis, analysis of variance and post hoc testing using the Tukey's honestly significant differences (HSD) test. If there was a significant interaction ($P < 0.05$), both the treatment groups and the interaction was reported. If there was no significant interaction, the differences between treatment diets and treatment environments were reported as the main effect comparisons. Results are reported as mean $\pm$ SEM for all measured variables.

affected relative organ weight ($P < 0.05$). There was a higher relative lung (0.87%) and heart (0.77%) weight for broiler chickens exposed to severe ammonia treatment than in non-ammonia exposed broiler chickens (0.57% and 0.55% respectively). Additionally, the right ventricle to total ventricle ratio for severely ammonia treated broiler chickens (0.32) was

greater than that of non-ammonia treated broiler (0.22), showing an increase in size of right ventricle and possibly pulmonary hypertension.

Diet and air quality are significantly interacting ($p < 0.05$) so that broilers fed control diets and exposed to high ammonia levels have higher relative lung weight (0.88%) and RV:TV ratio (0.32) than broilers fed Moringa oil (lung=0.86%; RV:TV =0.32). Additionally, broilers fed Moringa oil experienced lower relative liver (2.71%) and intestine weights (0.60%) than broilers fed control diets (2.73% and 0.67% respectively), indicating hepatic congestion and intestinal inflammation were lower than in control chickens when both groups were exposed to moderate concentrations of ammonia. This agrees with findings from [20] that noted that derivatives of Moringa enhance tissue integrity and modulate systemic inflammation by improving the antioxidant balance.

4.2 Kidney Function and Nitrogen Metabolism

With regard to elevating and increasing levels of ammonia, serum levels of creatinine and urea showed a statistically significant increase ($P < 0.05$) with increasing levels of ammonia (table three). This is indicative of renal stress and impaired nitrogen metabolism. Serum creatinine rose from 0.46 to 0.68 mg/dl ($P < 0.05$) and serum urea rose from 5.14 to 7.37 mg/dl ($P < 0.05$). When compared to non-supplemented birds, those supplemented with moringa oil had lower levels of creatinine (0.65 mg/dl vs 0.71 mg/dl) and urea (7.45 mg/dl vs 7.30 mg/dl).

findings were etched out when broilers facing oxidative stress and environmental

These results corroborate the findings of [21] that consumption of bioactive compounds from *Moringa oleifera* increased renal function by increasing animal detoxification and reducing renal oxidative stress. In conclusion, these data provide evidence that moringa oil can reduce nephrotoxicity associated with ammonia exposure through its antioxidant effects and greater metabolic efficiency.

4.3 Liver Enzymes and General Cellular Damage (AST, ALT, LDH)

Exposure to ammonia has significantly impaired activity of particular hepatic enzymes as well as compromised structural integrity of liver cells (table four). The increase in serum levels of AST from 236.19 IU/L to 334.27 IU/L (severe ammonia) at 21 days old; also, statistically significant increase noted when comparing ALT values at 9.89 IU/mL (non-ammonia) to 14.90 IU/mL (severe ammonia) represents hepatocyte injury. Furthermore, LDH was assessed as a marker for tissue damage and demonstrated increase from 249.22 mg/dl (non-ammonia) to 288.97 mg/dl (severe ammonia).

When compared to non-supplemented birds that were exposed to high ammonia levels, the birds that were supplemented with Moringa oil had lower serum levels of AST, ALT and LDH as compared with those birds that were not supplemented with Moringa oil. The mechanism of action of Moringa oil with regards to its protective properties on the liver may be related to its ability to limit oxidative damage and support the cellular structure of the hepatocytes. Similar

stress were supplemented with Moringa seeds or Moringa oil [22;23]

The increased concentrations of AST, ALT and LDH when exposed to ammonia may be attributed to damage to the mitochondria, inflammatory damage caused by oxidative stress; moreover, the phytochemicals present in Moringa (quercetin and phenolic acids) can enhance the activity of the endogenous antioxidant enzymes and stabilize the cellular membranes [21].

4.4 Lipid Metabolism (Cholesterol, Triglycerides, HDL, LDL)

Ammonia exposure resulted in severe alterations in lipid metabolism (table three). On Day 21, total cholesterol increased from 138.60 to 157.79 mg/dL and triglycerides increased from 60.99 to 65.35 mg/dL ($P < 0.05$) while HDL had a slight decrease (66.41 to 63.76 mg/dL) and LDL increased from 59.99 to 80.96 mg/dL.

Supplementation with Moringa oil significantly reduced this effect. Birds supplemented with Moringa oil had significantly lower LDL concentrations than birds fed the control diet ($P < 0.05$) and triglycerides (58.20 vs 64.53 mg/dL) compared to control birds (no supplemented). Similarly, Moringa supplemented birds consistently had lower LDH values compared to no supplemented birds. The reduction in LDL, elevation of HDL, and reduction of triglycerides in birds fed on Moringa could possibly be attributed to Moringa's polyunsaturated fatty acid content and plant sterol content, which enhance hepatic β -oxidation and regulate the synthesis of lipoproteins [23].

Consistent results were described by [22], who found that broilers fed Moringa seed cake exhibited elevated serum HDL and decreased serum LDL and triglycerides. Therefore, the findings from this study

suggest that Moringa oil promotes lipid metabolism, reduces the oxidative damage to cells [23].

The stress response to exposure to ammonia included multiple systems within the body (respiratory, hepatic, metabolic) demonstrated with the use of: elevated indices of the lung and heart, increases in transaminases and LDH activity; and disruption of the normal lipid profile. This suggests ammonia exposure will repeatedly, continuously induce an imbalance in metabolism and increase oxidative stress through poor ventilation in broilers.

4.5 Integrated Physiological Interpretation

The addition of *Moringa oleifera* oil at 1.0% of the diet improved many of the detrimental metabolic parameters associated with the changes caused by ammonia exposure; including: reduced transaminases and LDH activity; improved lipid metabolism; and reduced cardiopulmonary hypertrophy. Overall, this data suggests that *Moringa oleifera* oil has broad protective effects against oxidative stress as a result of ammonia exposure due to the polyphenolic and tocopherol fractions in Moringa, which can synergistically scavenge free radicals and increase the expression of antioxidant enzymes [23 21].

Table 2: Effects of dietary *Moringa oleifera* oil supplementation and ammonia exposure on relative organ weights (% liver, % lung, % heart, % intestine) and right ventricle to total ventricle ratio (RV:TV) in broiler chickens.

Treatments	% Liver		% Lung		% Heart		% Intestine		% RV:TV	
	14 D	21 D	14 D	21 D	14 D	21 D	4 D	21 D	14 D	21 D
	Diet									
Non Moringa Oil	3.15 a	2.72 a	0.81 a	0.69 a	0.66 a	0.66 a	5.03 a	4.43 a	0.21 a	0.26 a
Moringa Oil	3.32 a	2.69 a	0.80 a	0.71 a	0.65 a	0.63 a	4.90 a	4.10 a	0.22 a	0.26 a
Ammonia Levels										
Non-Ammonia	3.34 a	2.29 a	0.80 a	0.57 b	0.66 a	0.55 c	4.82 a	3.98 a	0.21 a	0.22 a
Moderate Ammonia	3.19 a	2.72 b	0.81 a	0.66 b	0.65 a	0.63 b	5.10 a	4.21 a	0.22 a	0.25 a
Severe Ammonia	3.18 a	3.11 c	0.79 a	0.87 a	0.64 a	0.77 a	4.99 a	4.60 a	0.21 a	0.32 a
Diet * Ammonia Levels										
Non Moringa oil Vs non-ammonia	3.24 a	2.25 b	0.83 a	0.54 b	0.68 a	0.58 cd	4.71 a	4.20 a	0.20 a	0.23 b
Non Moringa oil Vs moderate ammonia	3.13 a	2.73 ab	0.78 a	0.66 b	0.68 a	0.67 b	5.29 a	4.35 a	0.21 a	0.25 b
Non Moringa oil Vs Severe ammonia	3.09 a	3.17 a	0.78 a	0.88 a	0.64 a	0.75 a	5.09 a	4.75 a	0.21 a	0.32 a
Moringa oil Vs non-ammonia	3.44 a	2.32 b	0.77 a	0.60 b	0.65 a	0.52 d	4.93 a	3.77 a	0.22 a	0.22 b
Moringa oil Vs moderate ammonia	3.25 a	2.71 ab	0.85 a	0.66 b	0.64 a	0.60 bc	4.90 a	4.08 a	0.22 a	0.25 b
Moringa oil Vs Severe ammonia	3.27 a	3.06 a	0.79 a	0.86 a	0.65 a	0.78 a	4.88 a	4.45 a	0.22 a	0.32 a
a–c Means within the same column with different letters differ significantly (P < 0.05).										
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Table 3: Effects of dietary Moringa oleifera oil supplementation and ammonia exposure on kidney function indices and serum lipid profile (uric acid, creatinine, HDL, LDL, cholesterol, and triglycerides) in broiler chickens

Treatments	Uric acid mg/dL		Creatinine mg/dL		HDL mg/dL		LDL mg/dL		Cholesterol mg/dL		Triglyceride mg/dL	
	14 D	21 D	14 D	21 D	4 D	21 D	14 D	21 D	14 D	21 D	14 D	21 D
	Diet											
Non Moringa Oil	5.16 a	6.39 a	0.38 a	0.57 a	67.36 a	65.77 a	57.71 b	60.65 a	135.68 a	139.33 a	53.04 a	64.53 a
Moringa Oil	5.64 a	6.06 a	0.40 a	0.46 b	70.12 a	62.09 a	75.98 a	76.28 a	158.57 a	150.01 a	62.34 a	58.20 a
Ammonia Levels												
Non-Ammonia	5.19 a	5.14 b	0.53 a	0.46 b	65.10 a	66.41 a	74.9 a	59.99 b	152.35 a	138.60 a	61.75 a	60.99 a
Moderate Ammonia	5.53 a	6.16ab	0.32 b	0.41 b	75.80 a	61.63 a	58.96 b	64.44 b	144.96 a	137.62 a	50.99 a	57.75 a
Severe Ammonia	5.48 a	7.37a	0.33 b	0.68 a	65.32 a	63.76 a	66.67 ab	80.96 a	144.06 a	157.79 a	60.34 a	65.35 a
Diet * Ammonia Levels												
Non Moringa oil Vs non-ammonia	4.82 a	5.87 ab	0.52 a	0.49 abc	59.42 a	68.21 a	73.43 a	45.90 b	145.35 a	126.19 a	62.50 a	60.39 a
Non Moringa oil Vs moderate ammonia	5.01 a	6.02 ab	0.32 a	0.52 abc	75.12 a	62.03 a	41.78 b	59.79 b	126.24 a	135.26 a	46.69 a	67.19 a
Non Moringa oil Vs Severe ammonia	5.64 a	7.30 a	0.32 a	0.71 a	67.53 a	67.08 a	57.93 b	76.27 a	135.45 a	156.55 a	49.94 a	66.00 a
Moringa oil Vs non-ammonia	5.56 a	4.42 b	0.54 a	0.34 c	70.77 a	64.60 a	76.36 a	74.09 a	159.34 a	151.01 a	61.04 a	61.59 a

Moringa oil Vs moderate ammonia	6.04 a	6.30 ab	0.33 a	0.41 bc	76.47 a	61.22 a	76.16 a 69.09 ab	163.69 a	139.97 a	55.29 a	48.30 a
Moringa oil Vs Severe ammonia	5.33 a	7.45 a	0.34 a	0.65 ab	63.12 a	60.44 a	75.40 a 85.66 a	152.67 a	159.04 a	70.74 a	64.70 a
a–c Means within the same column with different letters differ significantly (P < 0.05).											

Table 4: Effects of dietary *Moringa oleifera* oil supplementation and ammonia exposure on serum liver enzymes and cellular damage markers (AST, ALT, and LDH) in broiler chickens

Treatments	AST IU/L		ALT IU/L		LDH mg/dL	
	14 D	21 D	14 D	21 D	14 D	21 D
	Diet					
Non Moringa Oil	215.81 a	265.04a	11.28 a	12.89 a	245.95 a	270.71 a
Moringa Oil	210.62 a	262.02a	10.91 a	12.63 a	259.23 a	275.09 a
	Ammonia Levels					
Non-Ammonia	201.07a	36.19b	9.71 a	9.89 b	236.89 a	249.22 a
Moderate Ammonia	230.22a	220.13b	10.89 a	13.49 a	269.15 a	280.53 a
Severe Ammonia	208.34a	334.27a	12.70 a	14.90 a	251.72 a	288.97 a
	Diet * Ammonia Levels					
Non Moringa oil Vs non-ammonia	189.01 a	228.51 b	9.62 a	8.52 v	229.99 a	237.89 a
Non Moringa oil Vs moderate ammonia	242.07 a	220.99 b	12.99 a	14.07 ab	254.34 a	278.84 a
Non Moringa oil Vs Severe ammonia	216.33 a	345.62 a	11.24 a	16.10 a	253.51 a	295.42 a
Moringa oil Vs non-ammonia	213.14 a	243.87 ab	9.80 a	11.27 ab	243.79 a	260.55 a
Moringa oil Vs moderate ammonia	218.37 a	219.280 b	8.78 a	12.92 ab	283.96 a	282.21 a
Moringa oil Vs Severe ammonia	200.34 a	322.93 ab	14.16 a	13.71 ab	249.92 a	282.53 a

a–c Means within the same column with different letters differ significantly ($P < 0.05$).

In terms of production, *Moringa oleifera* oil is a potential feed additive that is sustainable, will provide benefits to ensure improved broiler resilience to the environmental stresses associated with high

levels of ammonia, and moderate the physiological stress associated with high levels of ammonia, thereby improving the animal's welfare and overall production efficiency.

5. Conclusion

In conclusion, ammonia caused a combined stress response involving respiratory, hepatic and metabolic systems as demonstrated by an increase in the indices of both the lungs and the heart; increases in the activity of transaminases and LDH; and disruption of lipid profiles. These findings demonstrate that high ammonia levels in air creates oxidative stress and metabolic imbalance in broilers.

The administration of *Moringa oleifera* oil at 1.0% of their diet alleviated several of these negative responses such as decreased activity of transaminases and LDH, improved lipid metabolism, and decreased cardiopulmonary hypertrophy reflecting a wide range of protective effects against oxidative stress caused by ammonia.

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