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Growth Performance and Haematological Responses of Starter Broiler Chickens to Graded Levels of Lime (Citrus aurantiifolia) Juice in Drinking Water

By

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Abstract

The use of natural feed additives is gaining prominence as an alternative to antibiotic growth promoters in poultry production. This study evaluated the effects of lime (Citrus aurantiifolia) juice supplementation in drinking water on growth performance and haematological indices of broiler chickens. A total of 120 unsexed Anak broiler chicks were randomly assigned to four treatments with three replicates of 10 birds each. Treatments consisted of lime juice inclusion at 0 (control), 10, 15, and 20 mL/L of drinking water. Birds were fed standard broiler starter diets for four weeks. Proximate analysis of lime juice revealed appreciable concentrations of vitamin A (1.0 mg), vitamin C (32.1 mg), calcium (14.3 mg), and phosphorus (13.2 mg), indicating its potential as a bioactive supplement. Growth performance improved significantly ($p < 0.05$) in lime-supplemented groups, with the 20 mL/L inclusion yielding the highest final body weight (456.22 g), average daily gain (16.29 g), and feed intake (1268.01 g), though the feed conversion ratio was optimal at higher inclusion levels compared to moderate levels. Haematological profiles showed enhanced haemoglobin concentration and mean cell haemoglobin concentration in supplemented groups, particularly at 20 mL/L, suggesting improved oxygen-carrying capacity. Eosinophil counts declined with lime inclusion, while lymphocyte counts were highest at 15 ml/L, reflecting positive immune modulation. Overall, lime juice supplementation improved growth, nutrient utilisation, and haematological responses, highlighting its potential as a cost-effective, natural alternative to synthetic growth promoters in broiler production systems.

Keywords: *broiler chickens, lime juice, growth performance, haematology, natural feed additives.*

Introduction

Broiler chickens are among the world's most important sources of animal protein, alongside pork and fish, due to their relatively short production cycle and high feed conversion efficiency (Ravindran, 2013). However, their productivity depends greatly on efficient feed utilization and maintenance of gut health. In this regard, dietary strategies that promote gastrointestinal health while improving growth performance are essential for sustainable poultry production.

One promising natural additive is lime (Citrus aurantiifolia) juice, which is rich in organic acids, particularly citric and ascorbic acids, as well as bioactive compounds such as flavonoids, carotenoids, and limonoids (Dwyer et al., 2005; Ogunlade et al., 2019). These bioactive constituents exhibit antimicrobial, antioxidative, and growth-promoting properties, making lime a potential alternative to synthetic feed additives.

Organic acids are well established as effective feed or water additives in poultry nutrition. They inhibit acid-intolerant pathogenic bacteria such as *Escherichia coli*, *Salmonella* spp., and *Campylobacter* spp., thereby enhancing gut microbial balance (Dibner & Buttin, 2002; Ricke, 2013). By lowering gastrointestinal pH, organic acids create unfavourable conditions for pathogens, stimulate the secretion of pancreatic enzymes, and improve protein and mineral digestibility (Mellor, 2009). Previous studies have shown that supplementation with organic acids in drinking water improves body weight gain, feed conversion ratio, and

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immune response in broilers (Abdel-Fattah et al., 2008; Haque et al., 2010).

The practicality of water acidification in poultry production is noteworthy, as water consumption generally exceeds feed intake, ensuring more consistent delivery of additives (Cornelison et al., 2005). This approach is particularly important in tropical climates, where high ambient temperatures can compromise feed intake but not water consumption (Olukosi & Dono, 2014).

Traditionally, antibiotics were incorporated into poultry diets as growth promoters. However, the global concern over antimicrobial resistance, coupled with increasing restrictions on antibiotic use, has intensified the search for safe and natural alternatives (Castanon, 2007; Dibner & Richards, 2005). Organic acids, phytobiotics, and plant-derived compounds have been identified as viable candidates due to their antimicrobial, immunomodulatory, and antioxidative properties (Windisch et al., 2008).

Despite the commercial availability of synthetic acidifiers, their cost often limits adoption by smallholder poultry farmers in developing countries (Ahsan et al., 2016). Citrus fruits, particularly lime, provide an affordable and readily accessible source of natural acidifiers. Lime juice contains citric and ascorbic acids in concentrations sufficient to exert positive effects on poultry health and performance (Ogunlade et al., 2019). Moreover, its antioxidative properties may further enhance immunity and reduce oxidative stress in broilers reared under tropical conditions.

Given these attributes, lime juice supplementation in broiler drinking water represents a farmer-friendly, sustainable, and climate-smart feeding strategy. It has the potential to improve growth performance, enhance nutrient utilisation, and strengthen immunity without the drawbacks associated with synthetic additives or antibiotics.

Therefore, this study investigates the growth performance and haematological responses of starter broiler chickens administered graded levels of lime juice in drinking water, to establish lime juice as a natural, cost-effective alternative to conventional acidifiers in poultry production.

Materials and Methods

Study Location

The experiment was conducted at the Poultry Unit of the Research Farms, Faculty of Agricultural Sciences, Nnamdi Azikiwe University, Awka, Anambra State, Nigeria. The site lies within the forest vegetation zone of southeastern Nigeria, at latitude 6.2437°N and longitude 7.1219°E, with an elevation of 15 ft (75–77 m) above sea level. The area is characterised by a humid tropical climate, with a mean annual rainfall of approximately 1,037 mm, an average daily temperature of 34.7 °C, and a relative humidity of 83% (Google Earth, 2018). These climatic conditions make the region suitable for intensive poultry production but also expose birds to environmental stressors such as heat stress, which may affect growth performance and immune response.

Processing of Lime Juice

Fresh lime fruits (*Citrus aurantiifolia*) were purchased from a local market in Awka. Fruits were carefully sorted to remove damaged or diseased ones, washed thoroughly with clean water to eliminate surface contaminants, and cut into transverse sections using sterile stainless-steel knives. Juice was extracted manually by squeezing, filtered through a clean muslin cloth to remove seeds and coarse pulp particles, and collected in sterile glass containers. The juice was stored in airtight bottles at refrigeration temperature (4 °C) until use to preserve its bioactive constituents, particularly citric acid, ascorbic acid, and flavonoids (Dwyer, Peterson, & Bhagwat, 2005; Ogunlade, Oladipo, & Olayemi, 2019). This fresh extract served as the natural acidifier additive in the experiment.

Experimental Birds and Management

A total of 120 unsexed Anak broiler chicks, one day old, were procured from FIDAN Chicks Farms, Ibadan, Nigeria. The Anak strain is a fast-growing commercial broiler breed widely used in Nigeria due to its high feed efficiency and carcass yield (Adebiyi et al., 2019). Upon arrival, chicks were brooded for two weeks under standard management practices. The pens were cleaned, disinfected, and fumigated prior to stocking. Wooden floor pens were prepared with a 7-cm layer of wood shavings as litter material. Heating was provided using coal pots with temperature monitored daily to maintain the recommended brooding range (32–34 °C in week 1, gradually reduced to 26 °C by week 3) (Afolayan, Afolayan, & Adelowo, 2013).

Feed and drinking water were supplied *ad libitum*. Routine management practices such as vaccination (against Newcastle disease and infectious bursal disease) and medication were observed following standard poultry health protocols (OIE, 2018).

Experimental Design and Treatments

The birds were randomly allotted to four dietary treatments in a completely randomized design (CRD). Each treatment consisted of 30 chicks, replicated three times with 10 chicks per replicate. The treatments were defined by graded levels of lime juice added to drinking water:

- **T1 (Control):** 0 ml/L (plain water)
- **T2:** 10 ml/L lime juice
- **T3:** 15 ml/L lime juice
- **T4:** 20 ml/L lime juice

The inclusion levels were chosen based on earlier reports suggesting beneficial effects of organic acidifiers within similar dosage ranges (Abdel-Fattah, El-Sanhoury, El-Mednay, & Abdel-Azeem, 2008; Haque, Chowdhury, Islam, & Akbar, 2010).

Experimental Diets

Experimental diets were formulated according to the nutrient requirements of broiler starter chickens as recommended by the NRC (1994). The gross composition of the starter diet is presented in Table 1, while the proximate composition is shown in Table 2. The formulation was designed to provide

*Corresponding Author: **Onunkwo, D. N**

adequate protein, energy, and essential amino acids (lysine and methionine) to support rapid early growth.

Table 1. Gross composition of experimental starter diet (kg/100 kg)

Ingredient	Inclusion (kg)
Maize	55.00
Soybean meal (45% CP)	30.00
Fish meal (65% CP)	3.00
Groundnut cake	5.75
Wheat offal	2.50
Bone meal	2.00
Limestone	1.00
Lysine	0.20
Methionine	0.20
Premix*	0.25
Salt	0.10
Total	**100.00**

*Vitamin–mineral premix supplied per kg diet: Vitamin A, 10,000 IU; Vitamin D3, 2,500 IU; Vitamin E, 40 mg; Vitamin K3, 2 mg; Vitamin B1, 2 mg; Vitamin B2, 6 mg; Niacin, 20 mg; Pantothenic acid, 10 mg; Vitamin B12, 0.02 mg; Folic acid, 0.5 mg; Biotin, 0.08 mg; Mn, 80 mg; Zn, 60 mg; Fe, 40 mg; Cu, 8 mg; I, 1 mg; Se, 0.2 mg.

Table 2. Proximate composition of experimental starter diet

Nutrient parameter	Starter diet (%)
Crude protein	21.00
Ether extract (fat & oil)	6.00
Crude fibre	5.00
Calcium	1.00
Available phosphorus	0.45
Lysine	1.00
Methionine	0.50
Sodium chloride (NaCl)	0.30
Metabolizable energy (kcal/kg)	2,900
Net weight (kg/bag)	25

Data Collection

The body weight of chicks was recorded weekly using a digital weighing scale to calculate weight gain. Feed intake was determined daily as the difference between feed offered and feed residue. Feed conversion ratio (FCR) was calculated as:

FCR=Feed intake (g)/Body weight gain (g)

At the end of the 4-week experimental period, blood samples were collected from two birds per replicate via the brachial (wing) vein using sterile 2 ml syringes. Approximately 3 ml of blood was collected per bird and transferred into EDTA-coated tubes to prevent coagulation.

Haematological Analysis

Haematological parameters determined included packed cell volume (PCV), red blood cell (RBC) count, white blood cell (WBC) count, haemoglobin concentration (Hb), mean corpuscular volume (MCV), and differential leukocyte counts (lymphocytes, heterophils, eosinophils, monocytes, and basophils). Standard laboratory methods recommended by the World Health Organization (WHO, 2013) were employed. PCV was determined using the microhaematocrit method, Hb concentration by the cyanmethemoglobin method, and RBC/WBC counts by the improved Neubauer hemocytometer technique. Differential leukocyte counts were carried out on blood smears stained with Wright's stain.

Statistical Analysis

The data generated were subjected to one-way analysis of variance (ANOVA) using SPSS software (Version 8.0; SPSS Inc., 2003). Treatment means were separated using Duncan's New Multiple Range Test (Duncan, 1955). Significance was considered at $p < 0.05$.

Results and Discussion

Proximate Analysis of Lime Juice

The proximate analysis of lime juice (Table 3) confirmed the presence of key micronutrients, notably vitamin A (1.0 mg), vitamin C (32.1 mg), calcium (14.3 mg), and phosphorus (13.2 mg). These nutrients are physiologically essential for poultry nutrition and overall performance. Vitamin C functions as an antioxidant and a stress alleviator, playing a pivotal role in enhancing immune function and mitigating oxidative stress in poultry exposed to environmental and metabolic stressors (Khalil et al., 2017). Calcium and phosphorus are fundamental for skeletal development, bone mineralisation, and metabolic processes, particularly in rapidly growing broiler chickens (Leeson & Summers, 2001). Vitamin A, though detected in lower concentration, remains critical for vision, epithelial cell integrity, and immune competence (McDowell, 2000).

The presence of these bioactive compounds indicates that lime juice is not only a potential acidifier but also a natural source of micronutrients capable of complementing poultry diets. Similar studies have reported that citrus extracts, rich in citric acid and flavonoids, provide antibacterial and immunostimulatory benefits that enhance growth performance in broilers (Oke et al., 2009; Mellor, 2009). Thus, the

*Corresponding Author: **Onunkwo, D. N**

inclusion of lime juice in drinking water presents a dual nutritional and functional benefit.

Table 3. Proximate analysis of lime juice

Parameters	Present (+); Absent (–)	Concentration
Vitamin A	+	1.0 mg
Vitamin C	+	32.1 mg
Calcium	+	14.3 mg
Phosphorus	+	13.2 mg

Growth Performance at the Starter Phase

The effect of lime juice supplementation on the growth performance of broiler chickens during the starter phase is presented in Table 4. Supplementation significantly influenced final body weight, average daily gain (ADG), feed intake, and water intake. Birds in the 20 ml/L lime juice group (T4) recorded the highest final body weight (456.22 g), ADG (16.29 g), and total feed intake (1268.01 g), compared to the control (434.67 g final weight and 15.52 g ADG). These results suggest that lime juice supplementation supports growth performance, consistent with earlier reports that organic acids enhance gut health, nutrient absorption, and feed efficiency (Dibner & Buttin, 2002; Ricke, 2003).

Interestingly, the feed conversion ratio (FCR) did not follow the same trend. The poorest FCR was observed in the 10 mL/L group (2.91), whereas the control and higher lime juice treatments recorded improved efficiency (2.33–2.57). This indicates that moderate inclusion may create suboptimal digestive or metabolic responses, while higher supplementation levels better optimise nutrient utilisation. Increased water intake in lime-supplemented groups is likely linked to elevated feed intake, as feed and water consumption are positively correlated in poultry (NRC, 1994).

Table 4. Effect of lime juice on growth performance of broilers at the starter phase

Parameters	T1	T2	T3	T4
Final weight (g)	434.67a ± 2.95	436.30a ± 6.73	429.35a ± 2.95	456.22b ± 3.49
Total feed intake (g)	1016.88a ± 22.78	1174.23b ± 31.28	1198.90b ± 66.08	1268.01b ± 27.75
ADG (g)	15.52a ± 0.01	15.58a ± 0.02	15.33a ± 0.01	16.29b ± 0.01
FCR	2.33a ± 0.58	2.91b ± 0.11	2.57a ± 0.17	2.57a ± 0.62
Water intake (ml)	14237.67a ± 342.71	18158.00b ± 123.48	19082.00b ± 409.23	19104.00b ± 899.87

Values with different superscripts differ significantly ($p < 0.05$).

These findings demonstrate that lime juice inclusion stimulates feed and water consumption, thereby promoting growth. Previous studies have attributed such improvements to citric acid's ability to lower gut pH, suppress harmful pathogens, and create an optimal environment for nutrient digestion (Abdel-Fattah, El-Sanhoury, El-Mednay, & Abdel-Azeem, 2008).

Haematological Responses at the Starter Phase

Haematological parameters are key indicators of the physiological and health status of birds. Results presented in Table 6 revealed significant treatment effects. Haemoglobin concentration increased progressively with lime juice supplementation, peaking at 9.03 g/100 mL in T4 compared to 7.00 g/100 mL in the control. Similarly, mean cell haemoglobin concentration (MCHC) was markedly higher in T4 (9.37%) compared to other groups. These findings indicate improved oxygen-carrying capacity and metabolic efficiency, in agreement with Rahmani and Speer (2005), who reported that organic acids enhance erythropoiesis and nutrient utilisation.

Eosinophil counts declined substantially in lime-treated groups, with complete absence in T3. This reduction may signify decreased allergic or parasitic responses, suggesting a potential role of lime juice in modulating immune responses. Conversely, lymphocyte counts peaked in T3 (5.00%), pointing towards an immune-boosting effect at moderate lime inclusion. Similar immune-enhancing effects have been documented with phytogenic additives rich in flavonoids and organic acids (Mansoub, Myandoab, & Chekani-Azar, 2011).

Table 6. Haematology of broiler chickens supplemented with lime juice at the starter phase

Parameters	T1	T2	T3	T4
Hb (g/100 mL)	7.00a ± 0.57	7.93a ± 0.87	8.07a ± 3.03	9.03a ± 1.33
RBC ($\times 10^3$/mm^3)	11.33a ± 0.88	13.00a ± 2.52	10.27a ± 1.46	10.80a ± 1.33
WBC ($\times 10^3$/mm^3)	11.33a ± 0.88	9.33a ± 0.67	8.33a ± 0.33	11.33a ± 1.53
MCHC (%)	4.33a ± 0.88	5.33ab ± 1.45	5.00ab ± 1.53	9.37b ± 1.45
Eosinophils ($\times 10^3$/μL)	6.00c ± 1.16	3.00b ± 1.00	0.00a ± 0.00	1.33ab ± 0.88
Lymphocytes (%)	2.00a ± 1.16	2.00a ± 1.53	5.00a ± 2.00	2.67a ± 1.76

Values with different superscripts differ significantly ($p < 0.05$).

Overall Implications

Collectively, these results demonstrate that lime juice supplementation in drinking water can enhance both growth performance and haematological indices in broiler chickens.

*Corresponding Author: **Onunkwo, D. N**

The improvements observed are attributable to the presence of natural organic acids (citric and ascorbic acids), flavonoids, and minerals, which enhance gut health, nutrient assimilation, and immune modulation. These findings corroborate previous studies supporting phytogenic feed additives and natural acidifiers as viable alternatives to antibiotic growth promoters (Mellor, 2009; Mansoub et al., 2011).

Thus, lime juice inclusion in broiler diets offers a cost-effective, locally available, and safe strategy for improving productivity, reducing disease risk, and promoting sustainable poultry production in tropical environments.

Conclusion and Recommendation

The results of the starter phase demonstrate the potential of lime (Citrus aurantiifolia) juice as a natural growth promoter in broiler chickens. Supplementation with lime juice improved key performance indicators, including feed intake, final body weight, average daily gain, feed conversion ratio, and water intake. Among the treatment levels, the 20 mL/L inclusion consistently produced the best outcomes, likely due to the optimal supply of citric and ascorbic acids required to enhance nutrient digestion and utilisation. These findings corroborate earlier reports that organic acids such as citric acid stimulate feed intake, improve digestibility, and consequently enhance body weight gain in broilers (Paul et al., 2007).

Haematological results at the starter phase also showed positive responses to lime juice supplementation, with increases in haemoglobin concentration, lymphocyte counts, and mean cell haemoglobin concentration, indicating improved oxygen transport, immune modulation, and overall physiological resilience. However, packed cell volume (2.03–3.33%) remained below the standard reference range of 35.9–41.0% (Wikivet, 2018), suggesting that lime juice supplementation may improve some blood indices without fully restoring others to optimal levels. Nevertheless, the haematological improvements recorded reflect better health and potential resistance to stress and disease, in agreement with earlier studies highlighting the diagnostic and ecological importance of blood indices in poultry (Onyeyili et al., 1992; Togun et al., 2007; Ovuru, 2004; Mmereole, 2008; Isaac et al., 2013).

Recommendation

Based on the findings from the starter phase, it is recommended that 20 ml of lime juice per litre of drinking water be used for broiler chickens to optimise growth performance and enhance haematological status. Further investigations should explore long-term effects and assess interactions with other natural feed additives to strengthen sustainable poultry production.

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