MODULATORY EFFECTS OF ORGANICALLY (VINEGAR) ACIDIFIED DRINKING WATER ON SERUM AND MEDULLARY MINERAL PROFILES OF BROILER CHICKENS

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ABSTRACT
This study evaluated the serum and medullary bone mineral profiles of broiler chickens supplemented [A1]with drinking water modified by organic vinegar at pH levels of 7, 6, 5, and 4.5. The results indicated alteration of some minerals concentration in both serum and bones. In the serum, levels of calcium were better off in water of pH level of 6 with means of (9.0±0.29 mg/dL. However, chloride was altered in this treatment (91.0±10.0 mmol/L). Bone zinc concentrations were highest in chickens receiving acidified water to a pH of 4.5 with a zinc content of 3.3±0.13mg/L, copper 0.21±0.02 mg/L, manganese 0.32±0.01 mg/Calcium 74.0±0.01 mg/L and magnesium 3.6±0.03 mg/L, and all were statistically significant (P<0.05). These findings suggest that water acidified with vinegar has more advantages in terms of improving minerals' absorption and the bone mass in case pH is low enough; hence, it may be considered as a technique for improving skeletal system structure and overall mineral economy in broilers.
Keywords: Broiler chickens, Serum minerals, Vinegar, Water acidification, Skeletal minerals.

INTRODUCTION

Optimal mineral utilization and skeletal health are essential for poultry production. Acidification of drinking water using organic acids, such as vinegar, has been shown to enhance nutrient absorption and gut health in poultry (El-Saadony et al., 2022). However, its influence on serum and skeletal mineral composition remains understudied. This study investigated the modulatory effects of vinegar-acidified drinking water on the mineral profiles of broiler chickens.

MATERIALS AND METHODS

Experimental Site

The study was conducted at the Teaching and Research Farm of the Faculty of Agriculture, Federal University Dutse, Jigawa State, Nigeria. Dutse is located at latitude 11.760°N and longitude 9.341°E. The area experiences an average annual temperature of approximately 25°C, with temperatures ranging from 21°C in the coldest month to above 35°C in the hottest month. The total annual rainfall is around 600 mm (Ibrahim et al., 2012).

Management of Experimental Birds

One hundred and twenty (120) day-old Cobb 500 broiler chicks were allocated to four (4) dietary treatments, each replicated three times with 10 chicks per replicate in a completely randomized design (CRD). The birds were housed in deep litter pens and managed with all necessary routine management practices and routine vaccinations. Standard brooding practices were followed during the first week, including the provision of heat, light, and a starter diet.

Diet

A nutritionally balanced commercial diet was used throughout the experiment. The proximate composition of the diet included: Crude protein: [22, 20 %], Metabolizable energy: [2950, 3050 kcal/kg] for starter (21 days) and finisher (14 days) feeding respectively. Feed and water were provided *ad libitum*.

Treatment and Experimental Design

The experiment utilized a completely randomized design (CRD) with four treatments, each representing a specific water acidification level. The treatments were as follows: T1, T2, T3, and T4 at 7.0, 6.0, 5.0 and 4.5 pH, respectively

Water Acidification

Water was acidified using vinegar (acetic acid), and the pH levels were adjusted using a pH meter (Model: SARTORIUS- PB-10 BASIC BENCH TOP pH METER[A2], Germany) prior to distribution. The water was freshly prepared each day and supplied in calibrated drinkers.

Mineral Profile Assessment
Serum Mineral Composition
At the end of the experiment, approximately 3–5 mL of blood was drawn from the brachial vein of three randomly selected broilers per replicate using sterile syringes. Blood was transferred into plain tubes and allowed to clot at room temperature for 30 minutes. The samples were centrifuged at 3,000 rpm for 10 minutes, and the resulting serum was harvested and stored at ice temperature until analysis. Serum sodium, chloride, potassium, calcium, and phosphorus concentrations were determined using an automated spectrophotometric method.

Medullary Mineral Composition
For medullary bone analysis, the femur and tibia bones of three randomly selected broilers per replicate were collected, cleaned of adhering tissue, and ashed at 550°C for 12 hours (Li et al., 2020). The mineral composition of the ash, including zinc (Zn), copper (Cu), manganese (Mn), iron (Fe), magnesium (Mg), potassium (K), phosphorus (P), calcium (Ca), and sodium (Na), was analyzed using atomic absorption spectrophotometry (PinAAcle 900H A Spectrometer, PerkinElmer Inc., USA). Phosphorous was determined by titration following acid digestion.

Statistical Analysis
Data were analyzed using one-way analysis of variance (ANOVA) in SPSS (version 23.0) statistical software. Treatment means were compared using Tukey's Honest Significant Difference (HSD) test at 5% significance level.

RESULTS AND DISCUSSION
Serum Mineral Composition
The results presented in Table 1 show the effects of organically acidified drinking water on serum mineral composition of broiler chickens. Serum Na, K, and P were not significantly (P>0.05) affected by acidification. However, Ca and Chloride were significantly (P<0.05) affected by acidification. The observed increase in serum Ca at pH 6.0 compared to pH 7.0 (control) aligns with the submission of Adil et al. (2010), who reported that dietary acidifiers enhanced calcium solubility and absorption. Acidified environments increase the ionization of calcium salts, facilitating their uptake in the gastrointestinal tract. However, the decline in serum calcium at pH 4.5 may suggest an overly acidic environment that impairs gut enzyme activity or damages mucosal integrity, as reported by Ajith et al. (2019). This indicates that moderate acidification (pH 6.0) is optimal for calcium bioavailability. The Na) and K levels remained statistically (P>0.05) unchanged across treatments, which is in agreement with the findings of Zhang et al. (2022) who observed minimal effects of organic acids on serum electrolytes. For chloride, the significant (P<0.05) differences observed, with lower levels at pH 5.0, indicate a possible interaction with altered electrolyte balance under acidified conditions.

Table 1: Effects of organically (vinegar) acidified drinking water on serum mineral composition of broiler chickens

Parameters	Treatment				P-Value
	1 (pH 7.0)	2 (pH 6.0)	3 (pH 5.0)	4 (pH 4.5)	
Sodium (Mmol/L)	131.5 ± 2.5	135.5 ± 2.5	136.5 ± 7.5	135 ± 3	0.562
Chloride (Mmol/L)	89 ± 10^{ab}	91 ± 10^{a}	86.5 ± 1.5^{c}	89.5 ± 0.5^{a}	0.005
Potassium (Mmol/L)	3.9 ± 1	3.6 ± 0.6	3.75 ± 0.15	3.95 ± 0.15	0.884
Calcium (mg/dL)	8.6 ± 0.1^{b}	9 ± 0.2^{a}	8.85 ± 0.15^{ab}	8.6 ± 0.1^{b}	0.022
Phosphorous (mg/dL)	0.85 ± 0.05	1.1 ± 0.1	1.05 ± 0.15	0.95 ± 0.05	0.054

Superscript abc = Means within the same row having different superscripts are significantly different (P<0.05),

Bone Mineral Composition
The results presented in Table 2 show the effects of organically acidified drinking water on medullary bone mineral composition of broiler chickens. All trace minerals except Fe were significantly (P<0.05) affected by acidification. However, Na, P, and K were also not significantly (P>0.05) affected by the level of acidification. The significant increase in Zn, Cu, Mn, Mg, and Ca levels in the bones at pH 4.5 signals the potential of vinegar as a bioavailability enhancer for trace minerals. These results are consistent with the work of Brenes et al. (2003), who emphasized that acidifiers improve the solubility and absorption of trace minerals by creating favorable ionic conditions in the gut. However, the decline in Zn and Cu levels at pH 5.0 compared to pH 4.5 contradicts findings by Zhang et al. (2022), who reported consistent improvements in mineral deposition with organic acids. This discrepancy could be attributed to differences in the acidifier type, concentration, or interaction with dietary components. Moreover,

the significant elevation of Mn and Mg at pH 4.5 could reflect enhanced intestinal absorption due to a more acidic gut environment facilitating their mobilization from feed matrices.

Table 2: Effects of organically (vinegar) acidified drinking water on medullary (femur and tibia) bone mineral composition of broiler chickens

parameters	Treatment				P-Value
	1 (pH 7.0)	2 (pH 6.0)	3 (pH 5.0)	4 (pH 4.5)	
Zinc (mg/L)	2.7 ± 0.08^{b}	2.9 ± 0.20^{b}	2.6 ± 0.13^{c}	3.3 ± 0.13^{a}	0.002
Copper (mg/L)	0.12 ± 0.01^{b}	0.12 ± 0.01^{b}	0.10 ± 0.01^{c}	0.21 ± 0.02^{a}	0.001
Manganese (mg/L)	0.24 ± 0.02^{b}	0.24 ± 0.02^{b}	0.22 ± 0.01^{b}	0.32 ± 0.01^{a}	0.001
Iron (mg/L)	3.9 ± 0.54	4.3 ± 0.71	3.4 ± 0.35	4.4 ± 0.54	0.194
Magnesium (mg/L)	3.4 ± 0.01^{b}	3.5 ± 0.05^{b}	3.4 ± 0.06^{b}	3.6 ± 0.03a	0.006
Potassium (mg/L)	96 ± 11	110 ± 14	110 ± 16	120 ± 6.6	0.196
Phosphorous (mg/L)	290 ± 21	310 ± 17	270 ± 23	290 ± 43	0.371
Calcium (mg/L)	73 ± 0.11^{b}	74 ± 0.29^{b}	73 ± 0.18^{c}	74 ± 0.01^{a}	0.001
Sodium (mg/L)	192 ± 8.3	201 ± 1.5	220 ± 30	210 ± 17	0.313

Superscript abc = Means within the same row having different superscripts are significantly different (P<0.05).

CONCLUSION

The findings conclude that medium acidification (pH 6.0) enhances serum mineral levels, while more acidic conditions (pH 4.5) boost trace mineral deposition in bones. These findings have practical implications for broiler production, particularly in environments with suboptimal mineral content in water or feed.

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