MITIGATING HYDROCARBON-INDUCED HAEMATOLOGICAL AND INFLAMMATORY STRESS IN CHICKENS USING AQUEOUS GINGER EXTRACT: A COMPARATIVE STUDY ACROSS THREE BREEDS

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ABSTRACT
This study investigated the impact on haematological response due to hydrocarbon exposure and the mitigating effects of ginger aqueous extract (GAE) supplementation in three chicken breeds: FUNAAB Alpha (FAC), Agrited broilers (RB) and Nigerian light local chicken ecotype (NDC). Using a 3 x 3 factorial design, 60 birds per breed were assigned to three treatment groups: T1(control, no pollutant exposure or GAE), T2 (pollutant exposure without GAE), and T3 (pollutant exposure with GAE at 14mL/L water). The birds were exposed to crude oil fumes on alternate days for five weeks. Pollutant exposure (T2) significantly reduced packed cell volume, haemoglobin, and red blood cell counts, while increasing erythrocyte sedimentation rate, white blood cell counts, platelets, heterocyte-lymphocyte ratios and interleukin-6 levels ($p<0.05$). Supplementation with GAE (T3) effectively alleviated these adverse effects. Although breed-specific variations in most haematological indices were non-existent, NDC showed higher interleukin-6 levels ($p<0.05$) indicating a heightened inflammatory response. RB and NDC exhibited greater resilience to pollutant stress compared to FAC; however, FAC response was greatly enhanced in T3. Supplementation with GAE effectively mitigated the adverse haematological and inflammatory effects induced by hydrocarbon exposure in chickens, with breed-specific responses influencing resilience under the polluted conditions.

Keywords: Chickens, Ginger, Haematological indices, Hydrocarbon pollutants, Inflammatory markers, Breed-specific responses

INTRODUCTION
Anthropogenic activities such as crude oil exploration and gas flaring have led to degradation of numerous ecosystems in oil exploration regions like the Niger Delta region of Nigeria. Hydrocarbon pollutants a major consequence of petrochemical activities pose a significant threat to animals by disrupting homeostasis and compromising their health (Sanderfoot and Holloway, 2017). Stress response, to xenobiotic factors like hydrocarbon pollutants, in birds can easily be assessed by evaluating various blood indices (Amakiri and Owen, 2010). Oleforuh-Okoleh et al. (2023) in their review discussed the possible role of dietary antioxidants in enhancing poultry's tolerance to hydrocarbon pollutants. Ginger (*Zingiberofficinale*) is rich in bioactive compounds - gingerols, shogaols and zingerone – possess antioxidant and anti-inflammatory properties, and has shown potentials in combating oxidative damage induced by environmental stressors (Alexandra and Alexandra, 2022). Genetic factors could play a major role in the biochemical mechanism/pathway thereby influencing the degree of response to exposure (Bianchini and Morrissey, 2020). In other words, to inform sustainable management practices in crude oil exploration and polluted regions, it is imperative to consider genetic diversity and their varied response when assessing the impact of environmental pollutants to poultry health and productivity. The study investigated the haematological and inflammatory responses of difference chicken breeds reared in a simulated hydrocarbon-polluted environment and the mitigating effect of dietary ginger supplementation.

MATERIALS AND METHODS
The study spanned for five weeks at the Department of Animal Science, Teaching and Research Farm, Rivers State University, Nkpolu-Oroworukwo, Port Harcourt, Nigeria. A total of 180-day-old chicks from three chicken breeds (60 birds/breed): the Nigerian light local chicken ecotype (NDC), FUNAAB Alpha chickens (FAC), and Agrited Ross 308 broilers (RB), were used for the study.
The NDC group was derived from fertile eggs obtained from chickens raised in crude oil-polluted communities in Rivers State (Eleme, Okrika, and Gokana), while the FAC and RB groups were procured from the Federal University of Agriculture, Abeokuta, and Agrited Farms, Ibadan, respectively.

At five weeks of age, the chicks were randomly assigned into three treatment groups: T1, T2, and T3, with four replicates per treatment in a 3 x 3 factorial design. T1 which served as the control group was housed separately (100 meters away) from the pollutant source/exposed group, with no exposure to pollutants or ginger aqueous extract (GAE). Birds in T2 and T3 were reared under controlled exposure to fumes from combustion of 1L of Bonny light crude oil within their poultry house. This was done on alternate days over a 35-day period. Whereas the T2 group received no GAE, the T3 group was supplemented with GAE (14mL/L water), prepared according to Oleforuh-Okoleh*et al.* (2015). All treatment groups were fed *ad libitum* with a finisher diet (19% crude protein and 3150 ME kcal - Happy Chicken Elite) and provided with clean drinking water. Necessary bio security measures and vaccinations against prevalent diseases in the region (Newcastle disease, gumboro, coccidiosis, and fowl typhoid) were strictly implemented. At the end of the exposure period, three birds were randomly selected from each replicate/treatment group and blood samples were collected from the brachial vein (5 mL per bird) into EDTA-coated vials. Samples were immediately stored in a cold-chain box at 4°C and subsequently analysed for key haematogical indices, including packed cell volume (PCV), haemoglobin (Hb) concentration, and red blood cell (RBC) count while erythrocyte sedimentation rate (ESR), white blood cell (WBC) count, heterophil-to-lymphocyte (H/L) ratio, platelet (PLT) count, and interleukin-6 (IL-6) levels were quantified as inflammatory biomarkers. Statistical analyses were performed using the General Linear Model (GLM) procedure in IBM SPSS (Version 25), and separation of means were conducted using Tukey's test at statistical significance set at p < 0.05.

RESULTS AND DISCUSSION

The haematological responses of the three chicken breeds (FAC, RB and NDC) under hydrocarbon exposure and ginger supplementation (GAE) are presented in Table 1.

Table 1: Haematological and inflammatory response of three chicken breeds exposed to hydrocarbon-pollutants and ginger supplementation[1]

Breed	TrT	PCV (%)	HB (g/dl)	RBC (10^6/µl)	ESR (mm/hr)	WBC (10^3/µl)	PLT (10^3/µl)	H/L	IL-6 (pg/ml)
FAC	T1	30.38^a	9.63^a	4.20^a	2.02^b	40.75^b	198.50^b	0.53^b	25.22^c
	T2	26.90^b	7.40^c	3.55^b	5.55^a	64.13^a	278.00^a	1.14^b	34.76^b
	T3	29.90^{ab}	9.64^a	4.17^a	1.84^b	25.88^c	231.63^{ab}	0.47^b	24.23^c
RB	T1	30.35^a	9.88^a	4.23^a	2.67^b	32.50^{bc}	172.88^b	0.53^b	35.12^b
	T2	27.90^b	8.19^b	4.06^a	3.32^b	48.38^b	230.00^b	1.07^b	36.52^b
	T3	28.93^{ab}	9.91^a	4.16^a	2.54^b	30.75^{bc}	191.13^b	0.73^{ab}	30.90^{bc}
NDC	T1	29.85^{ab}	8.68^b	4.11^a	2.52^b	36.13^b	185.13^b	0.59^b	53.02^a
	T2	27.50^b	7.69^c	3.91^a	4.48^a	61.75^a	273.75^a	1.68^a	51.28^a
	T3	29.30^{ab}	9.76^a	4.11^a	2.46^b	44.75^b	203.00^b	0.91^{ab}	33.01^b
SEM		0.41	0.15	0.05	0.17	2.54	7.51	0.08	1.29

[ab]*Means on the same column with different superscripts are significantly different (p<0.05)*
[1]*Aqueous ginger extract* T1 Control group, T2: Exposure without ginger supplement; T3: Exposure with ginger supplement FAC: FUUNAB Apha, RB: ROSS 308 broilers, NDC: Nigerian light local chicken ecotype (NDC)
PCV – Packed Cell Volume; Hb – Haemoglobin; RBC- Red Blood Cell; ESR – Erythrocyte sedimentation rate; WBC White blood cell; HET- Heterophil, MON- Monocyte, H- Heterophil; L= Lymphocyte; IL-6 – Interleukin-6

Exposure to pollutants (T2) caused a significant reduction (p<0.05) in PCV, Hb, and RBC levels across all breeds, which is indicative of anaemia (Nwaogu and Onyeze, 2014; Juyeon and Hyun-Jin, 2024, Oleforuh-Okoleh et al., 2024), with FAC showing the highest drop in PCV (26.90%) and Hb (7.40g/dl). Amakiri and Owen (2010) made similar observations in broiler chickens exposed to simulated crude petroleum emissions. Restored erythrocyte indices levels due to GAE supplementation (T3) across all breeds reflects its protective role on erythrocytes by enhanced erythropoiesis or less erythrocyte destruction, perhaps attributable to its anti-inflammatory and antioxidant properties (Masuda, 2004; Alexandra and Alexandra, 2022). T2 also increased (p<0.05) immune and inflammatory markers (ESR, WBC counts, H/L ratios, PLT counts and IL-6 levels) across all breeds suggesting heightened inflammation and stress. Breed-specific responses were observed with FAC under T2 showing spiked ESR and WBC evidence of increased inflammation and immune activation. Furthermore, PLT count, H/L ratios and IL-6 peaked significantly and the NDC was the most affected. Conversely, GAE supplementation (T3) reduced the inflammation biomarkers substantially across all breeds, restoring them close to the control (T1), with FAC and RB demonstrating better recovery compared to NDC. The inflammatory response due to exposure which was greatly reduced by nutritional modulation with GAE indicates the benefits from antioxidant therapies (Alexandra and Alexandra, 2022; Cocou et al., 2023), buttressing the opinion of Nitish et al. (2024) of ginger's potential to modulate inflammatory cytokine production.

CONCLUSION

From our findings, we conclude that exposure to hydrocarbon pollutants poses a significant threat to poultry health by inducing anaemia; inflammation and stress, with varying breed susceptibilities. Furthermore, nutritional intervention using ginger aqueous extract in mitigating pollutant-induced stress is a promising strategy towards enhancing poultry resilience in polluted regions, particularly in resilient breeds like Ross 308 broiler and FUNAAB Alpha chickens. Therefore, breed-specific responses should constitute a fundamental component of poultry genetic improvement programmes.

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