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Genetic Variation in Heat Tolerance and Immunocompetence of Chickens Raised in Nigeria

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ABSTRACT

Background and Objective: Knowledge of genetic variations in heat tolerance and immunocompetence between different strains is imperative in genetic selection and enhancement of natural disease resistance in the tropics. Hence, this study compared Nera Black, Dominant Blue and Nigerian indigenous chicken strains. **Materials and Methods:** Heat tolerance was evaluated by measuring the pulse rate, respiratory rate, rectal temperature and heat stress index of the birds. Blood analysis was carried out to determine the blood differentials of the birds. A haemagglutination inhibition (HA/HI) test on blood samples from the birds before and after antigenic challenge with newcastle disease virus (NDV) was used to evaluate immunocompetence. **Results:** Nera Black had the highest means for heat tolerance traits while the Nigerian indigenous had the least. Sex significantly ($p<0.05$) influenced all heat tolerance traits except rectal temperature. Genotype had a significant ($p<0.05$) effect on the blood parameters with mean values for heterophil percentage, heterophil/lymphocyte ratio and monocyte percentage lowest for the Nigerian indigenous. The Nigerian indigenous had the highest postvaccination titer mean. **Conclusion:** From this study, the Nigerian indigenous chickens had higher heat tolerance and immunocompetence and were therefore better adapted to the tropics than the exotic birds studied.

KEYWORDS

Haemaglutination inhibition, heat stress, newcastle disease virus, Nigerian indigenous chickens, tropics, blood differentials, immune response, antibody titer

INTRODUCTION

Poultry is one of the largest agricultural businesses in Nigeria, which has undergone an enormous expansion and development over time[1,2]. However, heat stress is one of the most serious factors affecting overall poultry production in the tropics and diseases are big causes of loss in the poultry industry[3,4].

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High environmental temperatures have deleterious effects on poultry, reducing the rate of growth, feed intake and nutrient digestibility, increasing mortality and reducing immunity[5-8]. The coordination of all the systems under thermal stress is different between species and also between breeds[9], thereby providing a possibility to select for heat tolerance[10].

In the same vein, knowledge of differences in disease resistance between different strains may be valuable in genetic selection programs[11]. Characterization and evaluation of immune parameters in various genotypes can help in enhancing natural disease resistance in tropical and subtropical environments[12].

This study was a comparative analysis of three strains of chickens in Nigeria in terms of their abilities to tolerate heat and provide natural immunity against the Newcastle disease virus.

MATERIALS AND METHODS

Experimental birds: A total of 96 birds were used for the experiment. Day-old chicks of the Nigerian indigenous chicken were obtained from the crossing of the parent stock of pure naked neck, pure normal-feathered and pure frizzle-feathered birds maintained at the Poultry Breeding Unit of the University of Agriculture, Abeokuta, while the day-old chicks of Nera Black and Dominant Blue were purchased from a commercial parent-stock breeding farm. The chicks were wing-tagged right from the hatchery. The experiment lasted between June and December, 2009.

Experimental location: The fieldwork was conducted at the Poultry Breeding Unit of the Teaching and Research Farm of the Federal University of Agriculture, Abeokuta (FUNAAB), Nigeria. Blood analysis was carried out at the Microbiology Laboratory of the College of Veterinary Medicine, FUNAAB.

Management and feeding: All chicks were subjected to the same controlled environment with conventional ventilation. Wood shaving litter was used at 3-5 cm thickness. Indoor ambient temperature was started at 34°C and then gradually decreased till the end of brooding.

At the brooder phase (0-6 weeks), the chick starter ration containing 21% crude protein and 10.88 MJ kg^{-1} metabolisable energy was fed to the chicks while at the grower phase (7-12 weeks), growers mash containing 15% crude protein and 10.46 MJ kg^{-1} metabolisable energy was fed to them.

Data collection

Heat tolerance traits: Rectal temperature was measured by inserting a clinical thermometer into the vent of each bird for 1 min and then taking the reading. To get the pulse rate, a stethoscope was placed under the wing vein to count the number of pulses for 15 sec. The obtained value was then multiplied by 4 to get beats/min. Respiratory rate was determined by placing the fingertips under the wing vein and counting the number of beats per minute using a stopwatch[13]. The measurements were done weekly for 20 weeks. The readings were taken twice, the averages were computed and recorded.

Heat stress index: The heat stress index was evaluated using the relationship between pulse rate and respiratory rate, with their normal averages as shown[14]:

$$H = \frac{AR}{AP} \times \frac{NP}{NR}$$

Where:

H = Heat stress index
AR = Average respiratory rate value
AP = Average pulse value
NP = Normal pulse rate value
NR = Normal respiratory rate

Blood parameters for heat tolerance: At 17 weeks of age, about 5 mL of blood was collected from each of the birds from the three strains, through the wing vein, 2 mL of which was dispensed into a clean Bijou bottle containing an anticoagulant (Ethylenediaminetetraacetic acid (EDTA)) and labeled accordingly. The un-coagulated blood was used to determine heterophils, lymphocytes, monocytes, basophils and eosinophils counts, using standard laboratory procedures. The heterophil/lymphocyte ratio was calculated from their counts. Serum was obtained from the remaining 3 mL of blood, from which potassium and sodium were also determined.

Blood parameters for immune response: At 17 weeks of age, blood samples were taken from the three strains. Newcastle disease vaccine (NDV) was then administered to the birds the same day. As 3 weeks later, blood samples were collected from the birds again. Both the pre-vaccination blood samples and the post-vaccination samples were analyzed for the HA/HI antibody titer.

Statistical analyses: Heat tolerance traits and blood parameters measured involved both sexes, male and female. The statistical model that was used therefore accommodated the effects of sex, genotype and their interaction. The model was as follows:

$$Y_{ijk} = \mu+G_i+X_j+(GX)_{ij}+E_{ijk}$$

Where:

Y_{ijk} = Observed value of the measurable traits of the jth sex on the ith genotype
μ = Overall mean
G_i = Effect of the ith genotype (i = 1,2,3)
X_j = Effect of the jth sex (j = 1,2)
$(GX)_{ij}$ = Effect of the interaction of the ith genotype and the jth sex
E_{ijk} = Error independently and identically distributed as normal with mean zero and constant variance

The data were analyzed using the General Linear Model of SAS 9.0 while Duncan's Multiple Range Test was used for separating the means[15]. The results are presented as Means±Standard Error (SE), at a 5% level of significance.

RESULTS AND DISCUSSION

The Nigerian indigenous strain had the least rectal temperature while the Nera Black had the highest (Table 1). The observed significant differences in rectal temperature agreed with the report by Finch[16] that there were notable differences between breeds in their abilities to regulate rectal temperature at normal environmental conditions. It was, however, observed that the rectal temperatures were within a specific range (39.41-39.98°C) among all three strains. Heat production is affected by body weight, specie/breed, production, feed intake, feed quality and activity/exercise[7,17].

The highest mean values for respiratory rate were observed among the Nera Black chickens, with the peak at the 12th week. As Robert[18] pointed out, the size of the animal affects the respiratory rate. As ambient temperature increases, the autonomic nervous system of birds triggers increased heartbeat (tachycardia), accompanied by an increase in respiratory rates, which helps in maintaining their body temperature[3,8,19].

The Nigerian indigenous chickens had the lowest mean value for pulse rate throughout the period of the study. This could be attributed to the fact that Nigerian indigenous chickens have been reported to be well-adapted to tropical and subtropical environmental conditions due to the accumulation of genes for adaptability through natural selection[1]. Fayeye *et al.*[20] reported that the feather distribution gene and the feather structure gene, which are found among the local birds, were also associated with increased heat tolerance.

Table 1: Least squares mean and standard errors for heat tolerance traits as affected by the genotype of chickens

Age (weeks)	Genotype	Number	Rectal temperature (°C)	Pulse rate (beats min^{-1})	Respiratory rate (breaths min^{-1})	Heat stress index
1	Dom. Blue	29	40.11 ± 0.08^{b}	311.60 ± 2.8^{c}	39.53 ± 0.9^{a}	1.19 ± 0.02^{b}
	Nig. Indig.	32	39.41 ± 0.14^{c}	309.16 ± 1.6^{b}	35.76 ± 0.4^{b}	1.17 ± 0.01^{b}
	Nera Black	35	40.87 ± 0.07^{a}	317.90 ± 2.1^{a}	42.86 ± 0.4^{a}	1.22 ± 0.01^{a}
4	Dom. Blue	29	39.95 ± 0.09^{b}	307.93 ± 2.8^{c}	36.20 ± 0.6^{a}	1.19 ± 0.01^{a}
	Nig. Indig.	32	39.35 ± 0.18^{c}	303.42 ± 1.6^{b}	31.29 ± 0.3^{b}	1.16 ± 0.01^{b}
	Nera Black	35	40.24 ± 0.14^{a}	310.67 ± 1.2^{a}	38.24 ± 0.2^{a}	1.20 ± 0.01^{a}
8	Dom. Blue	29	40.02 ± 0.11^{b}	303.66 ± 2.7^{b}	35.07 ± 0.8^{a}	1.19 ± 0.03^{a}
	Nig. Indig.	32	39.40 ± 0.09^{b}	298.53 ± 1.9^{a}	31.47 ± 0.4^{b}	1.13 ± 0.02^{b}
	Nera Black	35	40.98 ± 0.07^{a}	308.19 ± 1.4^{a}	39.30 ± 0.3^{a}	1.16 ± 0.01^{ab}
12	Dom. Blue	29	40.08 ± 0.13^{b}	324.73 ± 2.9^{b}	38.13 ± 0.7^{a}	1.17 ± 0.04^{b}
	Nig. Indig.	32	39.42 ± 0.08^{b}	313.63 ± 1.8^{a}	34.66 ± 0.4^{a}	1.13 ± 0.01^{c}
	Nera Black	35	40.65 ± 0.14^{a}	339.07 ± 1.5^{ab}	45.01 ± 0.3^{a}	1.20 ± 0.01^{a}
16	Dom. Blue	29	39.99 ± 0.30^{b}	318.40 ± 2.8^{b}	36.03 ± 0.7^{a}	1.15 ± 0.03^{c}
	Nig. Indig.	32	39.31 ± 0.11^{c}	306.37 ± 1.5^{a}	30.92 ± 0.4^{b}	1.16 ± 0.01^{b}
	Nera Black	35	40.33 ± 0.08^{a}	325.65 ± 1.6^{a}	40.07 ± 0.2^{b}	1.22 ± 0.01^{a}
20	Dom. Blue	29	39.82 ± 0.16^{b}	314.67 ± 4.6^{b}	34.67 ± 0.7^{a}	1.15 ± 0.03^{b}
	Nig. Indig.	32	39.24 ± 0.07^{c}	308.00 ± 4.7^{a}	30.68 ± 0.4^{a}	1.10 ± 0.01^{c}
	Nera Black	35	40.18 ± 0.13^{a}	320.09 ± 2.3^{b}	38.63 ± 0.2^{a}	1.17 ± 0.01^{a}

[a,b,c]Means in the same column with different superscripts are significantly different ($p<0.05$)

Table 2: Least squares mean and standard errors for heat tolerance traits as affected by sex

Age (weeks)	Sex	Number	Rectal temperature (°C)	Pulse rate (beats min^{-1})	Respiratory rate (breaths min^{-1})	Heat Stress Index
1	Male	39	39.82 ± 0.16^{a}	308.11 ± 2.83^{b}	32.58 ± 0.90^{b}	1.18 ± 0.03^{b}
	Female	57	40.15 ± 0.10^{a}	$315.\ 89\pm1.58^{a}$	38.33 ± 0.3^{a}	1.22 ± 0.01^{a}
4	Male	39	39.75 ± 0.27^{a}	305.20 ± 1.19^{b}	30.20 ± 0.67^{b}	1.17 ± 0.02^{b}
	Female	57	40.02 ± 0.10^{a}	310.01 ± 1.20^{a}	36.78 ± 0.20^{a}	1.21 ± 0.01^{a}
8	Male	39	39.73 ± 0.12^{a}	298.35 ± 2.63^{b}	30.60 ± 0.54^{b}	1.14 ± 0.02^{a}
	Female	57	39.98 ± 0.05^{a}	309.34 ± 1.40^{a}	38.34 ± 0.26^{a}	1.15 ± 0.01^{a}
12	Male	39	39.88 ± 0.22^{a}	307.36 ± 1.69^{b}	34.78 ± 0.70^{b}	1.14 ± 0.02^{a}
	Female	57	40.31 ± 0.12^{a}	313.03 ± 1.32^{a}	39.45 ± 0.23^{a}	1.16 ± 0.01^{a}
16	Male	39	39.80 ± 0.09^{a}	304.40 ± 1.87^{b}	33.21 ± 0.09^{a}	1.15 ± 0.02^{a}
	Female	57	40.07 ± 0.06^{a}	310.23 ± 1.28^{a}	36.76 ± 0.22^{a}	1.19 ± 0.03^{a}
20	Male	39	39.75 ± 0.19^{a}	300.02 ± 2.43^{a}	30.54 ± 0.53^{b}	1.13 ± 0.02^{a}
	Female	57	39.94 ± 0.12^{a}	305.43 ± 1.80^{a}	34.45 ± 0.24^{a}	1.18 ± 0.01^{b}

[a,b]Means in the same age in the same column with different superscripts are significantly different ($p<0.05$)

The Nera Black had the highest means for all heat tolerance traits measured throughout the period of the experiment. This indicated a sign of susceptibility to heat stress[8]. The mean values for all the strains were high in the 1st week of life, however, the highest mean was at week 12. The fluctuation in pulse rate can be associated with factors like temperature, disease conditions or intense activity by the animal[21].

Heat stress index is defined as a function of the deviation of actual temperature from target environmental temperature and bird age[22]. The higher the index, the more stressed the bird[23]. The Nera Black had the highest mean value, making them the most stressed of the other genotypes while the Nigerian indigenous chickens had the least, making them the least stressed. The highest heat stress index was observed at week 1, which decreased with age, an indication of adaptation to tropical environment as they advanced in age. The observed pattern of heat tolerance traits among the chicken strains is similar to that reported by Lara and Rostagno[24].

Sex was found to have no significant effect on rectal temperature, though the males had lower values than the females throughout the experimental period (Table 2). However, there was a significant difference between sexes for pulse rate, respiratory rate and heat stress index. This also agreed with the report of the University of Illinois Extension aforementioned. Adedeji *et al.*[25] reported a significant ($p<0.05$) effect among the sexes of pure and crossbred chicken progenies in response to heat tolerance traits, with the females having higher responses compared to their male counterparts.

Table 3: Least squares mean and standard error for heat tolerance traits as affected by the interaction of genotype and sex of chickens

Age (weeks)	Gen×Sex	Rectal temperature (°C)	Pulse rate (beats min^{-1})	Resp rate (breaths min^{-1})	Heat stress index
	Dom. Blue male	39.68±0.28[bc]	306.87±0.12[ab]	32.13±0.27[bc]	1.17±0.08[b]
1	Dom. Blue female	40.22±0.32[ab]	316.22±0.85[ab]	36.86±0.17[ab]	1.22±0.07[a]
	Local male	39.43±0.21[bc]	301.08±0.14[b]	30.82±0.19[c]	1.16±0.02[b]
	Local female	39.97±0.23[bc]	310.94±0.17[ab]	33.04±0.13[bc]	1.18±0.03[b]
	Nera male	40.12±0.43[ab]	308.48±0.33[ab]	39.10±0.15[a]	1.16±0.08[b]
	Nera female	41.02±0.21[a]	326.38±0.08[a]	42.23±0.13[a]	1.15±0.07[b]
	Dom. Blue male	39.62±0.11[ab]	301.67±0.48[b]	31.23±0.61[bc]	1.18±0.09[a]
4	Dom. Blue female	40.18±0.18[ab]	318.79±0.22[a]	35.03±0.26[ab]	1.20±0.01[a]
	Local male	39.32±0.23[a]	296.75±0.27[c]	30.03±2.10[c]	1.13±0.06[b]
	Local female	39.78±0.11[b]	302.68±0.16[b]	33.57±0.21[b]	1.16±0.05[b]
	Nera male	40.32±0.12[a]	306.37±0.30[b]	33.45±0.26[b]	1.18±0.05[b]
	Nera female	40.89±0.19[a]	321.28±0.19[a]	39.10±0.24[a]	1.24±0.12[a]
	Dom. Blue male	39.58±0.27[b]	312.77±0.44[ab]	32.70±0.26[b]	1.13±0.18[b]
8	Dom. Blue female	40.26±0.31[ab]	318.49±0.72[a]	35.11±0.32[ab]	1.17±0.09[ab]
	Local male	39.42±0.21[b]	295.20±0.81[b]	29.75±0.21[c]	1.13±0.09[b]
	Local female	39.63±0.92[b]	309.02±0.29[ab]	31.88±0.20[b]	1.15±0.05[b]
	Nera male	40.14±0.12[ab]	311.18±0.46[a]	33.95±0.21[ab]	1.17±0.12[b]
	Nera female	41.32±0.12[a]	316.29±0.76[a]	34.52±0.22[ab]	1.23±0.20[a]
	Dom. Blue male	39.63±0.45[b]	307.83±0.65[c]	38.20±0.15[a]	1.16±0.14[b]
12	Dom. Blue female	40.18±0.42[ab]	315.87±0.25[b]	35.86±0.41[ab]	1.18±0.08[ab]
	Local male	39.37±1.23[bc]	305.82±0.76[c]	25.37±0.24[c]	1.11±0.09[b]
	Local female	39.59±0.18[b]	312.55±0.22[b]	32.18±0.18[bc]	1.14±0.12[b]
	Nera male	40.34±0.46[ab]	315.47±0.18[b]	32.32±0.11[bc]	1.18±0.14[ab]
	Nera female	40.93±0.48[a]	343.85±0.09[a]	38.88±0.24[a]	1.21±0.09[a]
	Dom. Blue male	39.45±0.87[ab]	308.47±0.20[b]	32.83±0.34[ab]	1.16±0.02[b]
16	Dom. Blue female	39.89±0.88[a]	319.91±0.33[ab]	34.33±0.50[ab]	1.19±0.07[ab]
	Local male	39.23±0.45[b]	300.16±0.25[c]	28.96±0.27[c]	1.14±0.09[b]
	Local female	39.38±0.34[b]	311.85±0.21[b]	30.73±0.18[b]	1.15±0.03[b]
	Nera male	39.97±0.45[ab]	319.58±0.32[ab]	33.97±0.22[ab]	1.21±0.03[a]
	Nera female	40.26±0.66[a]	328.66±0.16[a]	39.35±0.24[a]	1.23±0.11[a]
	Dom. Blue male	39.74±0.22[ab]	302.37±0.67[bc]	30.90±0.17[b]	1.14±0.19[b]
20	Dom. Blue female	40.02±0.19[a]	318.54±0.43[b]	37.50±0.48[a]	1.17±0.21[ab]
	Local male	39.23±0.34[b]	298.50±0.24[c]	29.33±0.27[b]	1.09±0.05[ab]
	Local female	39.45±0.23[b]	308.31±0.19[bc]	33.98±0.18[ab]	1.12±0.17[b]
	Nera male	39.87±0.32[ab]	308.22±0.54[bc]	32.70±0.17[ab]	1.16±0.09[ab]
	Nera female	40.45±0.67[a]	322.76±0.15[a]	37.67±0.17[a]	1.19±0.22[a]

[a,b,c]Means for the same ages in the same column with different superscripts are significantly different ($p<0.05$)

The sex by genotype interaction was significant on heat tolerance traits in agreement with the submission of Adedeji *et al.*[25], with the Nigerian indigenous males having the lowest means. The interaction of sex by genotype on rectal temperature (Table 3) brought out the effect that was masked when the sex effect alone was considered.

The means of blood parameters for the three strains were significantly different (Table 4). Lymphocyte and eosinophil counts were significantly higher in the local birds than in the other two strains. The ratio of heterophils to lymphocytes was also found to be significantly lower for the Nigerian indigenous chickens. It has been reported that differential leucocyte count and the H/L ratio are sensitive indicators of stress responses relevant to immune function[26,27]. High heterophil counts and the H/L ratio have been related to stress[28]. The above results, therefore, demonstrated higher heat tolerance in the Nigerian indigenous strain than the Nera Black and the Dominant Bluebirds in agreement with previous reports of the superiority of local strains in this regard[27]. According to Maxwell and Robertson[29], eosinophils disappeared from circulation and basophils increased in circulation during stress, particularly acute stress. The higher the eosinophils and the lower the basophils, the more heat tolerant the bird is.

Table 4: Least squares mean and standard errors of blood parameters as affected by the genotype of chicken

Genotype	K^+	Na^+	H (%)	L (%)	H/L	M (%)	B (%)	E (%)
Dom. Blue	4.1±0.07^a	133.5±1.76^a	28.8±0.88^a	64.4±0.15^b	0.44±0.05^b	2.33±0.33^a	2.32±0.02^a	3.09±0.10^b
Nig. Indig.	3.9±0.07^a	133.5±0.5^a	25.8±1.39^b	67.2±1.11^a	0.38±0.04^c	2.36±0.33^a	2.29±0.03^a	3.32±0.50^a
Nera Black	3.9±0.08^a	134.5±0.88^a	29.6±0.67^a	63.5±0.67^b	0.46±0.03^a	2.30±0.11^a	2.45±0.02^a	3.03±0.67^b

[a,b,c]Means in the same column with different superscripts are significantly different ($p<0.05$), K^+: Potassium ion concentration, Na^+: Sodium ion concentration, H (%): Heterophil percentage, L (%): Lymphocyte percentage, H/L: Heterophil/lymphocyte ratio, M (%): Monocyte percentage, B (%): Basophil percentage and E (%): Eosinophil percentage

Table 5: Least squares mean and standard errors for antibody titer as affected by the genotype of chickens

Genotype	Pre-vaccination titre (log 2)	Post-vaccination titre (log 2)
Dom. Blue	0.93±0.71^a	4.12±0.59^b
Nig. Indig.	1.50±0.51^a	4.50±0.89^a
Nera Black	0.87±0.46^a	3.92±0.77^b

[a,b]Means in the same column with different superscripts are significantly different ($p<0.05$)

The means of the pre-vaccination and post-vaccination antibody titers as affected by the genotype of chickens were presented in Table 5. The means of the pre-vaccination titer were not significantly different. The means of post-vaccination antibody titer were significantly higher for the Nigerian indigenous strain while Dominant Blue and Nera Black were not significantly different in means.

This showed that the Nigerian indigenous birds generated a higher immune response to the newcastle diseases virus than the other two strains. The local chickens had been said to possess genes that are believed to confer not only adaptability to the tropical climate but also disease resistance[26,30]. Alvarez *et al*.[31] stated that the indigenous naked neck and normal-feathered chickens seem to have better immune responses than the commercial chicken line. There are many such reports which indicate that, in general, rural chickens are resistant to many endemic diseases and stressful environments and survive better than commercial chickens under rural conditions[27,31,32]. The significant difference in the blood differentials did not only reveal heat tolerance level, but it was also a measure of immune function. Reports showed that leukocyte counts also had been used as a measure of immune function in studies on sexual selection in birds[32,33].

This study showed that differences in genotype accounted for variations in heat tolerance traits, rectal temperature, respiratory rate and pulse rate as well as variations in the blood parameters measured. It also revealed that the Nigerian indigenous local chickens have higher heat tolerance as well as a higher immune response to the Newcastle disease virus, thereby possessing greater adaptability for the tropical environment.

From this study, it can be recommended that the Nigerian indigenous chicken strains be used in the genetic improvement of heat tolerance and immunocompetence in commercial strains through cross-breeding. This will provide a low-cost solution that is easier to achieve in developing countries with hot climates and endemic diseases. Also, the indigenous birds can be developed into commercial lines by raising them intensively, thereby conserving the genes they possess as well as aiding future breeding endeavours.

A limitation of this study is the fact that it only compared three strains out of many and assessed resistance to only one disease out of several that are economically important in the tropics.

CONCLUSION

The major findings of this research are the superior performances of the Nigerian indigenous chickens in terms of heat tolerance and immunocompetence (especially against the newcastle disease virus). The Nigerian indigenous birds have not been given as much credit as they deserve. As observed from this study, they have an edge over the commercial strains studied when raised intensively in the tropics.

SIGNIFICANCE STATEMENT

It is essential to assess the adaptation of our local birds in comparison with the exotic raised within our climate. Hence this study compared the genetic variations in heat tolerance and immunocompetence of Nera Black, Dominant Blue and Nigerian indigenous chickens. The major findings of this research are the superior performances of the Nigerian indigenous chickens in terms of heat tolerance and immunocompetence (against the newcastle disease virus), which showed that they have an edge over the commercial strains studied when raised intensively in Nigeria and the tropics in general.

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