

TRANSFORMING GROWTH FACTOR-B (TGF- β 3) GENE VARIATIONS IN DUAL-PURPOSE CHICKEN STRAINS AND ITS ASSOCIATION WITH GROWTH TRAITS

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ABSTRACT

Studies on TGF- β 3 genes suggests that they are crucial for maintaining healthy tissues and development and are located on chicken chromosome 5 which are close to genes that might influence body weight and feed conversion ratio (FCR) in chickens. TGF- β 3 gene polymorphs in dual purpose chicken were investigated to understand their association with various chicken growth and morphological traits in three chicken populations. Methods: 450 chickens of 150 per strains were used, comprising of Noiler® (150), FUNAAB Alpha® (FA - 150), and Sasso® (150), reared from day old to 20 weeks of age. Growth and morphological traits such as body weight (BW), height at withers (HW), body length (BL), breast girth (BG), and wing length (WL) were measured from 4 weeks to 20 weeks of age at 4 weekly intervals. Genomic DNA was extracted for polymerase chain reaction (PCR) with primers of TGF β 3 gene. PCR amplicons were sequenced and genotyped using genotype by sequence method, with two SNP primers as two SNP genotypes were identified. The identified SNPs were on exon 3 at positions 7941745 and 7941722 of the TGF β 3 gene which were named 7941745A>C and 7941722A>T respectively. Chi-square results demonstrated that the population used was under Hardy-Weinberg equilibrium (HWE) for the two SNP genotypes, with 7941745A>C and 7941722A>T having a dominant allele A. Marker-trait association results revealed that 7941745A>C was not significantly associated ($P>0.05$) with breast girth, body length and wing length, however, there was a significant association ($P<0.05$) with body weight and height at withers, with strains Sasso and FA showing more association than Noiler. 7941722A>T had no significant association with all the growth traits except body weight ($P<0.05$) with strains Sasso and FA showing more association. Marker-trait association results suggest that AA and CC genotypes for SNPs 7941745A>C gene could be used as a genetic marker when improving body weight and size of the chicken strains under selection, whereas genotype AA and AT could be used in improving bodyweight only amongst the chicken strains studied. This research, therefore, suggests that the TGF- β 3 gene could be a candidate gene that significantly influences the body weight and height at withers of chicken, hence might be used as potential genetic markers in dual purpose chicken breeding.

Keywords: Chicken, TGF- β 3 gene, SNP genotype, Growth, Breeding

INTRODUCTION

Chickens raised for meat or eggs are bred to grow quickly and produce well. These traits, along with the makeup of the chicken's body (carcass traits), are influenced by many genes (Anh *et al.*, 2015). Scientists are still figuring out how these genes work together to control growth, which is a complex process involving hormones and the nervous system (Okafor *et al.*, 2021). Traditionally improving chicken growth, meat quality, and egg production has been challenging because these traits are influenced by many genes. But, new breeding techniques like marker-assisted selection can help. This method uses specific genes linked to these traits to predict a chicken's overall genetic potential for good performance (Li *et al.*, 2003). These special genes are like signposts pointing to the desirable qualities.

MATERIALS AND METHODS

This research was carried out at the Demonstration Farm of the Faculty of Agriculture, University of Port-Harcourt, Choba, Port-Harcourt, Nigeria, where the chicken strains – Funaab Alpha, Noiler and Sasso parents were reared. The eggs collected from the F1 generation of the chicken strains were hatched and raised from day old to 20 weeks of age, to conduct this experiment.

Experimental Birds and Procedure

A total of four hundred and fifty (450) day old chicks of both sexes comprising 150 Noiler, 150 Funaab Alpha and 150 Sasso strains with average of 60 males and 90 females respectively per strain were all hatched, and reared from day old till 20 weeks of age, from eggs of their parents reared in the farm. The three strains were developed independently of each other by the breeders of the different strain.

Phenotypic Data

Data on growth parameters were collected on a weekly basis while feed intake record was collected daily. Body weight was measured using electronic scale and measuring tape for the body measurements -breast girth, body length, wing length and height at withers. The body measurements were taken from day old to 20 weeks of age as follows:

- Breast girth (cm): This was taken as the circumference of the breast around the deepest region of the breast; measuring tape was used to take the reading.
- Body Length (cm): This was taken as the distance between the last cervical vertebral before the thoracic vertebra and the caudal vertebrae and were done with the use of a measuring tape.
- Wing length (cm): The length of the wing from the end of the wing to the bone that attaches the wing to the body was recorded as the wing length. Measurement was taken using the measuring tape.
- Height at withers (cm): This was taken as the neck base line to the foot and was done with the use of a measuring tape.

Genomic Data

Genomic data was collected from blood samples taken from each of 150 birds through the wing vein at 20 weeks of age, prepared and processed according to the procedure of (Okoro *et al.*, 2014). DNA amplicons were sent to the genomic lab at IITA for sequencing and further bioinformatics analysis.

Genomic analysis

Genotype and Allele frequencies

The SNP genotypes and allele frequencies were calculated by simple allele counting method (Chromas, 2018). The possible deviations of allele and genotype frequencies from the Hardy–Weinberg equilibrium was examined with Chromas Bioinformatics Software (Chromas, 2018) with Pearson's Chi-square test.

Genomic Association analysis

Fifty birds each from Noiler, FA and Sasso strains respectively with pedigree information of their parents were used for an association study of genetic markers with body weight and morphological traits. Genomic and the phenotypic data generated were subjected to Association analysis based on ANOVA using General Linear Model Procedure of SPSS software (SPSS, 2022), with fixed effect of Strain (L_i), fixed effects of SNP genotype (SG_j), random effect of family (F_k) and random error term e_{ijk} effects according to the model:

$$Y_{ijk} = \mu + L_i + SG_j + F_k + e_{ijk}$$

Where, Y_{ijk} is the response variable and μ is the population mean. Significant differences between least-squares means of the different genotypes were calculated using DuncanNew Multiple Range Test (SPSS, 2022). SNPs that deviated from the Hardy-Weinberg equilibrium based on significant Chi-square values were discarded. Also, SNPs with minor allele frequency <1% across all individuals were removed.

RESULTS AND DISCUSSION

Meanwhile, the height at withers parameter was significantly associated amongst all the strains for AA genotype, while amongst the CA genotypes, the FA strains were significantly higher than the N strains. Also, amongst the 7941722A>T SNP genotypes, only two were identified – AA and TA (Table 5), with body weight being the only trait associated. All the strains were amongst the genotypes segregated under AA, with FA being significantly more associated than the others. However, only S strain was segregated amongst the TA SNP genotypes being significantly higher amongst all the other strains in both AA and TA genotypes.

These results were similar to the findings of (Li *et al.*, 2003) and (Amirinia *et al.*, 2011) that showed significant association with growth traits of chicken. Hence selecting those genotypes for future

Table 5. Association of the polymorphism of TGF β 3 gene with growth traits of chicken strains studied

SNPs	Genotype	Strain	BW (g) Mean $\pm$ SEM	HW (cm) Mean $\pm$ SEM	BL (cm) Mean $\pm$ SEM	BG (cm) Mean $\pm$ SEM	WL (cm) Mean $\pm$ SEM
7941745A>C	AA	N	1477.0 $\pm$ 131.60 ^b	11.55 $\pm$ 0.70 ^{ab}	9.35 $\pm$ 0.47	10.95 $\pm$ 0.51	8.50 $\pm$ 0.36
		FA	1554.0 $\pm$ 83.23 ^b	10.86 $\pm$ 0.44 ^{ab}	8.86 $\pm$ 0.30	10.16 $\pm$ 0.32	7.94 $\pm$ 0.22
		S	2030.0 $\pm$ 186.10 ^a	13.40 $\pm$ 0.99 ^a	11.20 $\pm$ 0.66	10.80 $\pm$ 0.73	8.80 $\pm$ 0.50
	CA	N	1232.1 $\pm$ 107.45 ^b	9.77 $\pm$ 0.57 ^b	8.47 $\pm$ 0.38	9.67 $\pm$ 0.42	7.40 $\pm$ 0.29
		FA	1375.0 $\pm$ 93.05 ^b	11.88 $\pm$ 0.49 ^a	9.28 $\pm$ 0.33	8.98 $\pm$ 0.36	7.82 $\pm$ 0.25
		S	1330.0 $\pm$ 100.84 ^b	10.48 $\pm$ 0.48 ^a	8.82 $\pm$ 0.33	10.18 $\pm$ 0.34	7.84 $\pm$ 0.27
7941722A>T	AA	N	1611.0 $\pm$ 92.05 ^a	11.1 $\pm$ 0.44 ^a	8.93 $\pm$ 0.30	10.23 $\pm$ 0.31	7.95 $\pm$ 0.25
		FA	1324.0 $\pm$ 130.18 ^b	11.53 $\pm$ 0.62 ^a	9.17 $\pm$ 0.43	8.70 $\pm$ 0.44	7.83 $\pm$ 0.35
		S	1778.0 $\pm$ 159.44 ^a	13.15 $\pm$ 0.76 ^a	10.41 $\pm$ 0.53	10.32 $\pm$ 0.54	8.33 $\pm$ 0.43
	TA	S					

Means with different superscripts ^{abc} within the same column of a particular SNP is significantly different ($p < 0.05$). N = Noiler, FA = Funaab Alpha, S = Sasso, BW = Body weight, HW = Height at Withers, BL = Body length, BG = Body girth, WL = Wing length.

breeding for high growth rate in dual purpose chicken will achieve rapid progress in their growth for both meat and egg production. This is in line with the general ideas that the process of identifying the QTL responsible for the economic important traits in chicken will facilitate poultry breeding programs as molecular genetic information is an easier and faster required means to enhance genetic improvement among animal species (Amirinia *et al.*, 2011). Also (Jin *et al.*, 2013) had reported that the candidate gene approach is a very powerful method, needed to investigate genomic associations of gene polymorphisms with economically important traits in farm animals.

Growth and body composition and its final expression is the result of interaction among genetic, nutritional and environmental factors, which are a comprehensive reflection of development of various parts of the chicken body (Jin *et al.*, 2013). The current research findings will help the dual-purpose chicken breeders because this study of TGF- β 3 gene as a potential candidate gene of QTL that is useful in the selection of chicken will help for both in improving meat and egg production. Our findings shows that TGF β 3 SNP has significant ($p < 0.05$) effect on body weight and height at withers at 20 weeks of age which is also in-line with the findings of (Amirinia *et al.*, 2011; Jin *et al.*, 2013; Li *et al.*, 2003) who reported that SNP in TGF- β 3 gene significantly ($p < 0.1$) affected body weight of broilers at 6 weeks.

CONCLUSION

Marker Assisted Selection can be an ideal option to improve selection in dual purpose chicken production. The results from the current study indicates that a SNP marker in the TGF β 3 gene was associated with body composition traits (body weight and height at withers) in chickens growing to point of lay, and are therefore, a potential marker for molecular MAS program in commercial dual purpose chicken lines, which is suitable for rural chicken farmers that needs sustainable meat and egg production in the long run.

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