UAR Publisher

UAR Journal of Multidisciplinary Studies (UARJMS)

ISSN: 3049-4346 (Online) | Published by UAR Publisher

LINKAGE OF MYOSTATIN GENE POLYMORPHISMS WITH CARCASS PARAMETERS AND EFFECTS OF AMINO ACID CHANGE ON PROTEIN FUNCTION OF BROILER CHICKENS

Adewole,R.A.[1], Adedeji,T.A.[2], Emiola,I.A.[3] and Adebambo,A.O.[4,5]

[1] Department of Animal Science, P.M.B. 1601, Al-Hikmah University, Ilorin, Nigeria.

[2] Department of Animal Production and Health, P.M.B. 4000, Ladoke Akintola University of Technology, Ogbomoso, Nigeria.

[3] Department of Animal Nutrition and Biotechnology, P.M.B. 4000, Ladoke Akintola University of Technology, Ogbomoso, Nigeria.

[4] Department of Animal Breeding and Genetics, P.M.B. 2240, Federal University of Agriculture, Abeokuta, Nigeria.

[5] ACUTIG Laboratory Nigeria Limited, Abeokuta.

*Corresponding Author: **Adewole Risikat Adeola**

Abstract

This experiment was designed to assess the association of *Myostatin* gene exon 1 polymorphisms with carcass parameters and determine the effect of amino acid change on protein function of four commercial broilers; Arbor Acre, Arbor Acre Plus Cobb and Ross. Two-hundred-day-old chicks of mixed sexes were randomly assigned into five equal-sized cells, each containing forty birds with ten birds per strain. The carcass characteristics considered were eviscerated weight, breast weight and thigh weight. At 12th week, blood samples were collected via brachial vein of forty birds for DNA extraction. Amplicons were visualized on 1% agarose gel. The amplified products were sequenced and single nucleotide polymorphisms were identified using codon code aligner. The results revealed twenty-seven polymorphisms comprising of twenty-two transition and five transversion mutations. Polymorphisms within exon 1 of the *Myostatin* gene significantly ($P<0.05$) associated with carcass parameters in the four commercial broilers. In conclusion, Arbor Acre birds with adenine and thymine alleles should be selected and

Article DNA

Article Type:
Original research article

DOI:
10.5281/zenodo.17913156

Article History:
Received: 02-12-2025
Accepted: 08-12-2025
Published: 12-12-2025

Keywords:
Myostatin, Single nucleotide polymorphism, Carcass parameter, Amino acid, Broiler, Allele.

How to Cite

Adewole, R. A., Adedeji, T. A., Emiola, I. A. and Adebambo, A. O. (2025). Linkage of Myostatin gene polymorphisms with carcass parameters and effects of amino acid change on protein function of broiler chickens. *UAR Journal of Multidisciplinary Studies (UARJMS)*, 1(10): 1-18.

***Related declarations are provided in the final section of this article.*

reared for meat. Cobb and Arbor Acre Plus with adenine allele should be considered while birds possessing adenine and guanine alleles in Ross should be selected and raised for meat production. Non-synonymous substitution in exon 1 such as proline to arginine may possibly influence growth and skeletal muscle development in the population by enhancing nutrient delivery to muscles. Mutation changing from threonine to histidine may influence muscle growth because histidine is essential for muscle growth. Mutation of aspartic acid changing to arginine in is a missense mutation, which disrupts function of *Myostatin* leading to increased carcass weight, muscle growth and development. Therefore, Ross can be recommended having higher *Myostatin* adenine and guanine alleles with non-synonymous mutation of aspartic acid to arginine which displayed superior carcass parameters confirmed suitability for high-yield meat production.

Introduction

Broiler chicken production in Nigeria has grown substantially in response to rising meat consumption (Oke *et al.,* 2024) and the government's support for self-sufficiency in poultry production. The most commonly raised commercial broiler strains in Nigeria include Ross 308, Cobb 500, Arbor Acre and Arbor Acre Plus, each exhibiting discrete growth rates, feed efficiency and carcass traits (Sam and Okon, 2022; Okolo, 2023). These differences emphasize the need for genetic studies to optimize strain selection for enhanced meat production.

Carcass characteristics such as eviscerated weight, breast weight and thigh weight are the key factors of broiler meat yield and quality. These traits influence market value, consumers' preference and overall profitability in poultry production (Tahmasbi, 2024). Although, nutrition, management and environmental factors affect carcass traits however genetics plays an essential role in determining muscle development and distribution (Zerehdaran *et al*., 2004).

Skeletal muscle constitutes 30 to 40% of the total body mass of chicken therefore, it is the most important tissue in meat producing animal (Hakamata *et al.,* 2020). It serves as food with good source of protein for man, hence there is an urgent need to improve muscle growth (Burt, 2005). Identifying genetic markers associated with superior carcass traits is therefore crucial for advancing selective breeding programmes and improving broiler productivity.

Myostatin (*MSTN*), also called growth differentiation factor 8 (GDF-8) is a member of the transforming growth factor-beta super-family (TGF-β) and a negative regulator of muscle growth and development (Rasmussen, 2016). Several association studies have explored the association between *MSTN* gene polymorphisms and carcass traits in poultry, revealing notable correlations between specific genetic variations and improved growth performance (Sartika *et al*., 2011).

The exon 1 region of the *Myostatin* gene is important in genetic studies because it encodes key regulatory domains that influence muscle development. Mutations or polymorphisms within this exon can lead to functional alterations in *Myostatin* activity, potentially enhancing muscle growth and carcass yield (Ran *et al*., 2017). Birds carrying beneficial mutations showed increased pectoral muscle mass, likely due to decreased *Myostatin* activity, which allows enhanced hypertrophy of muscle fibres (Zerehdaran *et al.,* 2004; Sharma *et al.,* 2015). Examining exon 1 polymorphisms in different broiler strains can help detect genetic markers contributing to improved meat production thereby assisting breeding programmes aimed at advancing high-performance broilers.

Materials and Methods

Experimental Site

The experiment was conducted at the Poultry Unit of the Teaching and Research Farm of the Department of Animal Science, Al-Hikmah University, Igbaja Campus, Kwara State located approximately 56kms north-east of Ilorin and north-north-east of Ajase-Ipo, on Latitude 8°23'60"N and Longitude 4°53'60"E at an altitude of 394.97 meters and 1295.83 feet above sea level (Google, 2023). The molecular analysis was carried out at Inqaba Biotech West Africa Limited, Ibadan, Oyo State located on latitude 7°23'47"N and longitude 3°55'0"E (Google, 2023).

Experimental Animals

Two-hundred-day-old broiler chicks of mixed sex comprising of fifty chicks per strain; Arbor Acre, Cobb 500, Ross 308 and Arbor Acre Plus were purchased from reputable hatcheries. Birds were raised in a deep litter system which was divided into five cells containing forty birds per cell thereafter; the birds were weighed, leg banded individually and later wing tagged at 4 weeks for easy identification.

Brooding and Rearing Management

Prior to the arrival of the day-old chicks, the brooder pen was partitioned into five cells for easy management and were washed with Lysol® and fumigated with formaldehyde, lime and potassium permanganate. The dimension of each cell was 10m × 10m. Wood-shaving was spread on the floor of each cell and charcoal heat source was placed at the centre of each cell to provide warmth for the chicks with thermometer to monitor temperature. At the arrival of the chicks, they

were given multi vitamin drugs which served as an anti-stress and were leg banded individually for identification purpose. The chicks were brooded for two weeks. Adequate ventilation, medication and ad libitum feeding were provided. The chicks were duly vaccinated and antibiotics was administered as required. At four weeks, the leg bands were replaced with wing tags. The litter was changed at five days intervals to prevent accumulation of ammonia gas. All chicks remained on deep-litter till 12 weeks.

Experimental Feeds and Feeding

The birds were given *ad libitum* access to commercial feed and water throughout the experiment. All birds were placed on convectional broiler starter crumble containing 21.0% Crude Protein and 2900kcal/kg Metabolizable Energy for six weeks and finisher pellet containing 17.0% Crude Protein and 3000kcal/kg Metabolizable Energy till the end of the experiment.

Data Collection

Carcass Parameters Sampling

At 12^{th} week of age, 10 birds from each genotype were randomly selected and fasted for 12 hours before being slaughtered by neck decapitation which was performed humanely by severing the jugular veins with a sharp knife by a single swipe. The following parameters were determined.

Live Weight (g)

This was measured in grams as the weight of an individual bird before being slaughtered.

Bled Weight (g*)*

Bled weight was determined in grams with sensitive scale after slaughtering process has occurred and blood was fully drained.

Pluck Weight (g)

The pluck weight was determined in gram when the bird was defeathered.

Eviscerated Weight (g)

Eviscerated weight was calculated as the weight of the carcass after the heart, crop, pancreas, lungs, digestive and urogenital tracts had been removed and weighed.

Primal Cuts

The carcass was cut into different parts known as primal cut for breast weight and thigh weight were determined in grams using sensitive balance scale.

Molecular Analysis Procedure

At 12th week of age, 1ml of blood was collected from 40 selected birds for carcass characteristics comprising of 10 birds from each of Arbo Acres, Ross 308, Arbo Acres Plus and Cobb 500. Blood was collected with syringe via brachial vein into ethylene diamine tetra acetic acid (EDTA) bottles as described by Ayorinde *et al.* (2001) and Sewalem *et al.* (2002) for molecular analysis.

Extraction of DNA

DNA was extracted from whole blood sample collected with the use of commercially available Quick-DNA^{TM} Miniprep Kit manufactured by Zymo Research Epigenetics Company, USA following the manufacturer's procedure.

DNA Quantification

Extracted gDNA was quantified for purity and concentration using Nanodrop spectrometer. The integrity of the DNA was checked using gel electrophoresis by running Iμl of each gDNA sample on 1% Agarose gel at 120V for 20 minutes.

Primer

The *myostatin* exon 1 gene was amplified using the designed forward and reverse primer sequences as used by Tanjung *et al.* (2019).

- Forward: 5′-ATGCAAAAGCTAGCAGTCTATG-3′
- Reverse: 5′-ACTCCGTAGGCATTGTGATAAT-3′

Polymerase Chain Reaction (PCR*)*

The extracted DNA was amplified using Polymerase Chain Reaction (PCR). The amplification reaction was carried out in PCR tube using programmable thermocycler (Mastercycler pro by Eppendorf). Polymerase Chain Reaction amplification was performed in a volume of 25 μl

mixture containing 2 µl of DNA template, 1 µl of each forward and reverse primer, 12 µl of master mix and 9 µl of sterilized distilled water.

Quantification of PCR Products

The concentration of the purified PCR product was determined using UV spectrophotometry or fluorescent quantification

DNA Electrophoresis

Electrophoresis was carried out on 2 µl sample from PCR amplicon at 100 volts for 45 minutes in 1 % agarose gel in 1 × TAE buffer containing 1.0 µl stain of ethidium bromide. After electrophoresis, the gel was taken into gel documentation machine to view the bands using Ultra-violet illumination. Moreover, for quality check, the PCR products (amplicons) was visualized using Agarose gel electrophoresis for the *Myostatin* gene with concentrations of 2.0%.

DNA Sequencing

Polymerase Chain Reaction (PCR) products of the DNA fragment was sequenced unidirectionally with forward primer using the Big Dye version 3.1 sequencing. The sequence data was collected automatically on the ABI PRISM 3100 Genetic Analyser (Applied Bio-systems).

Editing and Analysis of Sequence

The sequence was aligned, trimmed and edited using Bioedit® software. The edited sequence was blasted (BLASTN) (http://www.ncbi.nlm.nih.gov/BLASTN/) against other sequences in the GenBank to ascertain the similarity and identity with other chicken sequences of *Myostatin* gene in the database. The edited sequences were further aligned using CLUSTALW for further analyses.

SNPs Identification

Codon Code Aligner was used for the detection of the Single Nucleotide Polymorphisms (SNPs) present in the chicken populations' sequences which were associated with reference sequence using CLUSTALW for alignment and DnaSP to confirm the SNPs.

Association Analysis

The relationship between SNP markers and eviscerated weight, breast weight and thigh weight were determined using a single-factor multivariate ANOVA. Means were separated using the Duncan Multiple Range Test procedure of SAS (2023) software.

The association model used is as follows:

$Y_i = \mu + G_i + \varepsilon_{ij}$

Where;

μ = Population mean;

G_i = Fixed effect of the i^{th} SNPs (1, 2…n)

ε_{ij}= Residual error.

Results and Discussion

The single nucleotide polymorphisms (SNPs) and their amino acid changes identified in exon 1 of *Myostatin* gene in Arbor Acre (AA) are presented in Table 1. Six SNPs were identified in the strain comprising of five transitions and one transversion. Polymorphisms 56A>G (singleton), 65A>G (parsimony), 200C>G (singleton) and 329C>T (parsimony) were specific to different loci in AA. Two SNPs were non-synonymous and had effect on the amino acid type, while four of the SNPs were synonymous and had no effect (neutral) on the amino acid type and the percentage similarity ranges from 99-100%.

Identified polymorphisms and effect of amino acid changes on protein function of Arbor Acre Plus (AAP) of *Myostatin* are presented in Table 2. Five SNPs were identified in the selected AAP strain comprising of three transitions and two transversions. Polymorphism 55A>G (singleton), 64A>G (parsimony), 199C>G (singleton) and 328T>C (singleton) were found in AAP loci. All the SNPs were synonymous and had no effect on the amino acid type and the percentage similarity was 100%.

The single nucleotide polymorphisms identified in exon 1 of *Myostatin* gene in Cobb 500 and the amino acid changes are presented in Table 3. Five SNPs were identified in the selected Cobb strain comprising of three transitions and two transversions. Polymorphism 55A>G (parsimony), 64A>G (singleton), 199C>G (singleton) and 328T>C (singleton) were found on Cobb loci. All

the SNPs were synonymous and had no effect on the amino acid type. The percentage similarity ranges from 99-100%.

The SNPs identified in exon 1 of *Myostatin* gene of Ross 308 (Ross) are presented in Table 4. Eleven SNPs were identified in the selected Ross strain comprising of seven transitions and four transversions. Polymorphism 3A>G (parsimony), 49A>G (parsimony), 50C>A (parsimony) 56A>G (singleton) 66A>G (parsimony) 201C>G (singleton) and 330T>C (singleton) were found on Ross loci. One of the SNPs was synonymous and had no effect on the amino acid type and other ten were non-synonymous and had effect on the amino acid. The percentage similarity was 99% all through.

Table 5 represents effect of single nucleotide polymorphism on carcass parameters in AA, AAP, Cobb and Ross chickens. There were significant ($P<0.05$) effects of SNPs on carcass parameters in all the strains. In AA and Ross strains, two SNPs (65A>G; 329C>T) and four SNPs (3A>G; 49A>G; 50A>C; 66A>G) associated better with carcass traits. However, one SNP each in AAP and Cobb associated with eviscerated, thigh and breast weights. Birds having A and T alleles had higher EW, TW and BW in AA while it was generally A allele birds in AAP and Cobb. However, higher carcass traits in Ross were observed in birds possessing either G or A allele.

Table 1: Identified polymorphism and effect of amino acids changes on protein function of arbor acre in *myostatin* gene (exon 1)

Genotype	SNPs	Mutation Type	SNPs Type	Amino Acid	Amino acid Type	Effect	Percentage Identity (%)
AE3F	56A>G	Transition	Singleton	CGG > CGG (Arginine > Arginine)	Synonymous	Neutral	99
AB2F	65A>G	Transition	Parsimony	CCG > CGG (Proline > Arginine)	Non-Synonymous	Effective	100
AC3M	65A>G	Transition	Parsimony	CCG > CGG (Proline > Arginine)	Non-Synonymous	Effective	100
AE3F	200C>G	Transversion	Singleton	GGC > GGC (Glycine > Glycine)	Synonymous	Neutral	99

AB2F	329C>T	Transition	Parsimony	ATG > ATG (Methionine > Methionine)	Synonymous	Neutral	100
AC3M	329C>T	Transition	Parsimony	ATG > ATG (Methionine > Methionine)	Synonymous	Neutral	100

A = Adenine, **G** = Guanine, **C** = Cytosine and **T** = Thymine

Table 2: Identified polymorphism and effect of amino acids changes on protein function of arbor acre plus on *myostatin* gene (exon 1)

Genotype	SNPs	Mutation	SNPs	Amino Acid	Amino acid	Effect	Percentage Identity (%)
		Type	Type		Type		
APC2M	55A>G	Transition	Singleton	GGT > GGT (Glycine > Glycine)	Synonymous	Neutral	100
APB8M	64A>G	Transition	Parsimony	GGT > GGT (Glycine > Glycine)	Synonymous	Neutral	100
APB9F	64A>G	Transition	Parsimony	GGT > GGT (Glycine > Glycine)	Synonymous	Neutral	100
APC2M	199C>G	Transversion	Singleton	GCT > GCT (Alanine > Alanine)	Synonymous	Neutral	100
APC2M	328T>C	Transversion	Singleton	CGA > CGA (Arginine > Arginine)	Synonymous	Neutral	100

A = Adenine, **G** = Guanine, **C** = Cytosine and **T** = Thymine

Table 3: Polymorphism identified and effect of amino acids changes on protein function of cobb on *myostatin* gene (exon 1)

Genotype	SNPs	Mutation	SNPs	Amino Acid	Amino acid	Effect	Percentage Identity (%)
		Type	Type		Type		
CA7M	55A>G	Transition	Parsimony	GGT > GGT (Glycine > Glycine)	Synonymous	Neutral	100
CB9M	55A>G	Transition	Parsimony	GGT > GGT (Glycine > Glycine)	Synonymous	Effective	99
CA7M	64G>A	Transition	Singleton	AGT > AGT (Serine > Serine)	Synonymous	Neutral	100
CA7M	199C>G	Transversion	Singleton	GCT > GCT (Alanine > Alanine)	Synonymous	Neutral	100
CA7M	328C>T	Transversion	Singleton	CGA > CGA (Arginine > Arginine)	Synonymous	Neutral	100

A = Adenine, G = Guanine, C = Cytosine and T = Thymine

Table 4: Polymorphism identified and effect of amino acids changes on protein function of ross on *myostatin* gene (exon 1)

Genotype	SNPs	Mutation	SNPs	Amino Acid	Amino acid	Effect	Percentage Identity (%)
		Type	Type		Type		
RC2F	3A>G	Transition	Parsimony	ATG > GTT (Methionine > Valine)	Non-Synonymous	Effective	99
RC9M	3A>G	Transition	Parsimony	ATG > GTT (Methionine > Valine)	Non-Synonymous	Effective	99
RB3M	49G>A	Transition	Parsimony	ACA > AGA (Threonine > Arginine)	Non-Synonymous	Effective	99
RC2F	49G>A	Transition	Parsimony	ACA > AGA ((Threonine > Arginine)	Non-Synonymous	Effective	99
RC2F	50A>C	Transversion	Parsimony Parsimony Singleton	ACA > CAT (Threonine > Histidine)	Non-Synonymous	Effective	99
RB3F	50A>C	Transversion	Parsimony Parsimony Singleton	ACA > CAT (Threonine > Histidine)	Non-Synonymous	Effective	99

RC2F	56A>G	Transition	Singleton	CGG > CGG (Arginine > Arginine)	Synonymous	Neutral	99
RC9M	66A>G	Transition		GTG > GGT (Valine > Glycine)	Non-Synonymous	Effective	99
RD3F	66A>G	Transition Transversion		GTG > GGT (Valine > Glycine)	Non-Synonymous	Effective	99
RC2F	201C>G	Transversion		CTG > GCT (Leucine > Alanine)	Non-Synonymous	Effective	99
RC2F	330T>C			GAT > CGA (Aspartic acid > Arginine)	Non-Synonymous	Effective	99

A = Adenine, G = Guanine, C = Cytosine and T = Thymine

Table 5: Effects of single nucleotide polymorphisms in exon 1 of MSTN gene on carcass parameters of four strains of broiler chickens

GEN SNPs	ALLELE	EW (g)	TW (g)	BW (g)
AA 65A>G	A	4344.00±493.37[a]	928.67±139.61[a]	1425.00±189.71[a]
	G	4133.00±555.00[b]	798.00±96.00[b]	1315.00±65.00[b]
329C>T	C	4235.00±555.00[b]	798.00±96.00[b]	1312.00±65.00[b]
	T	4343.00±493.00[a]	929.00±139.61[a]	1428.00±89.71[a]
AAP 64A>G	A	4228.67±288.71[a]	1152.00±119.24[a]	1248.00±100.47[a]
	G	4120.00±279.00[b]	1091.00±97.00[b]	1123.00±93.00[b]
COBB 55A>G	A	4434.00±479.02[a]	955.33±186.17[b]	1422.00±199.27[a]
	G	4251.00±439.00[b]	1077.00±165.00[a]	1144.00±110.00[b]
ROSS 3A>G	A	4319.33±500.54[b]	983.33±138.24[b]	1374.67±97.34[b]
	G	4718.00±786.00[a]	990.00±392.00[a]	1472.00±110.00[a]
49A>G	A	4626.00±694.00[a]	906.00±308.00[b]	1466.00±99.36[a]
	G	4380.67±561.85[b]	1039.33±187.52[a]	1378.67±104.00[b]
50A>C	A	4674.00±694.00[a]	1191.00±191.00[a]	1331.00±151.00[b]

	C	4380.67±561.85[b]	849.33±186.64[b]	1402.00±70.01[a]
66A>G	A	4365.33±478.01[b]	937.33±180.5[b]	1304.00±86.31[b]
	G	4649.00±855.00[a]	1059.00±323.00[a]	1478.00±40.00[a]

[ab] = Means occupying the same column having different superscripts are significantly different (P<0.05). **GEN** = Genotype, **EW** = Eviscerated Weight, **TW** = Thigh Weight, **BW** = Breast Weight, **A** = Adenine, **G** = Guanine, **C** = Cytosine and **T** = Thymine.

Discussion

In this study, multiple single nucleotide polymorphisms (SNPs) were identified in exon 1 of the *Myostatin* (*MSTN)* gene across four commercial broiler strains. These polymorphisms were evaluated for their association with carcass parameters specifically eviscerated weight, breast weight and thigh weight. However, the AA showed moderate but consistent performance, which may indicate a more balanced growth profile favouring long-term meat quality and adaptability as discussed by Sartika *et al*. (2011). The molecular analysis of the *MSTN* gene revealed single nucleotide polymorphisms within exon 1 and the association analysis demonstrated a statistically significant relationship between certain *MSTN* genotypes and carcass performance. These results support earlier findings by Zhang *et al.* (2018) and Ran *et al.* (2017) who recorded that *MSTN* exon 1 polymorphisms influence muscle growth and are associated with variations in carcass yield in broilers.

These inter-strain differences in SNP expression and carcass parameters also reflect the strain-specific effect of *MSTN* polymorphisms as previously emphasized by Fasiola *et al.* (2017) and Zhang *et al.* (2018). The presence of both shared and unique SNPs across strains suggested that *MSTN* regions are evolutionarily conserved and associated with general growth traits, others may be strain-specific and respond differently to local environmental and nutritional conditions. The observed variation in carcass parameters among the broiler strains are consistent with prior studies that emphasised strain-specific performance in terms of growth rate, feed conversion, and muscle yield (Adebambo *et al.*, 2010; Olawunmi, 2020).

A total of six SNPs detected in Arbor Acre with significant (P<0.05) associations observed at loci 65A>G and 329C>T, this is in agreement with the work of Zhang *et al. (*2010). Birds carrying the adenine allele at 65A>G exhibited higher eviscerated weight, thigh weight and

breast weight suggesting that this allele may increase muscle accretion. This is consistent with the findings of Zhang *et al.* (2018), who documented that specific *MSTN* mutations particularly in exon 1 could lead to decrease *Myostatin* activity and consequently increase muscle fibre hypertrophy. Similarly, Ran *et al.* (2017) demonstrated that *MSTN* suppression in broilers leads to enhanced muscle growth and carcass parameters.

Five SNPs were identified in the Arbor Acre Plus strain, with polymorphism at position 64A>G showing significant effects on carcass parameters which aligns with Zhang *et al.* (2010). The adenine nucleotide at this locus was associated with improved eviscerated, thigh and breast weights which supported previous studies of Oladokun *et al.* (2020) and Fasiola *et al.* (2017), who documented that Arbor Acres Plus exhibited better feed efficiency and breast muscle yield, possibly due to favourable genetic traits such as *MSTN* polymorphisms. Moreover, the present findings correspond with Bellinge *et al.* (2005) and Aviagen (2021) on strain-specific genetic architecture, which emphasizes that even closely related strains like AA and AAP can possess divergent growth-related alleles impacting performance outcomes under similar environmental conditions.

Five SNPs were discovered in Cobb with notable significant effect observed at the 55A>G locus. Broilers carrying the A allele had significantly higher eviscerated and breast weights. This observation was in line with studies by Zhang *et al.* (2018) and Adeyemo *et al.* (2019), who found that Cobb birds possess superior genetic potential for meat yield and feed efficiency. Ran *et al.* (2017) further supported these findings, stating that Cobb are known for outstanding breast-to-body weight ratio and early maturity, traits potentially regulated by *MSTN*-related pathways.

Ross had the highest number of SNPs (11) with significant effects at loci 3A>G, 49A>G, 50A>C and 66A>G which disagrees with the report of Aviagen (2021). However, these SNPs were strongly associated with higher eviscerated and breast weights, indicating the presence of advantageous alleles for muscle deposition. These findings conform with those of Zhou *et al.* (2006) and Olawunmi (2020), who highlighted that Ross consistently outperformed other strains in carcass parameters due to its genetic optimization for rapid growth and muscle yield. Ross is emphasised for its high breast muscle development and feed efficiency, attributes which may be influenced by the *MSTN* polymorphisms identified in this study (Aviagen, 2021).

When comparing the SNP distribution and trait performance across strains, Ross emerged as genetically superior in eviscerated and breast weights (4718.00±786.00g) and (1472.00±110.00g) respectively. This supports the narrative that this strain is better suited for intensive meat production due to their selection history and genetic makeup according to Olawunmi (2020) and Adebambo *et al.* (2010). Ross exhibiting superior eviscerated and breast weights also agree with previous research which indicated that the strain is genetically selected for rapid growth and high breast muscle yield according to Fasiola *et al.* (2017). These differences emphasize the important role of genetic background in shaping carcass traits and justify the importance of strain-specific breeding strategies.

Amino acids sequences in proteins are genetically predetermined as essential for production of adequate quantities of meat, milk and eggs (Iyakutye and Hollinshead, 2011). Codon usage profoundly influences gene expression and protein functionality across a spectrum of organisms. In the context of expressing heterologous genes, optimising the codons of the target gene to match the preferred codons of the host species can significantly improve gene expression efficiency. In this study, Proline changing to Arginine metabolism is one of the central pathways for the biosynthesis of the amino acids. Arginine, an essential amino acid for poultry, has been shown to directly or indirectly see to derivative molecules, alleviate oxidative stress, improve antioxidant capacity and attenuate the intestinal mucosa disruption (Fernanda *et al.,* 2020).

In this experiment, mutation that changed the methionine codon to a valine codon would result in the insertion of valine instead of methionine during protein synthesis. Valine is an essential amino acid that involves in maintaining higher reproductive performance in chicken and aids muscle tissue repair. When threonine changes to arginine; arginine is a dibasic amino acid (Rubin *et al.,* 2007) consisting of a linear chain of four carbon molecules and a distal complex guanidinium group, displaying resonance hybrid properties that impart the chemical properties of arginine (Khajali and Wideman, 2010). Arginine is an essential amino acid in poultry due to the absence of a functional urea cycle.

This experiment revealed a mutation changing threonine to histidine during protein translation in the chicken genome which is a specific type of genetic change called a missense mutation which occurs when a single nucleotide changes within a codon to alter the codon's meaning, causing it to code for a different amino acid. Histidine is a vital component of proteins, which are essential for growth, maintenance and repair of tissue. Histidine is also involved in the production of histamine which plays a role immune response of chicken. When valine is converted to glycine

through a single nucleotide substitution at the first position which is achieved by transition. Aspartic acid changing to arginine in exon 1, can lead to increased body and carcass weight as well as muscle growth. This occurs because *Myostatin* is a negative regulator of muscle growth and the mutation often a missense mutation, can disrupt its function, leading to more muscle development.

Conclusion and Recommendation

This study has clearly demonstrated that polymorphisms within exon 1 of the *Myostatin* (*MSTN*) gene significantly associated with eviscerated weight, chest weight and thigh weight in the four commercial broiler strains. The influence of *MSTN* polymorphisms on breast weight observed in this study aligned with the established function of *Myostatin* as a negative regulator of muscle growth. The detection of SNPs within the *MSTN* gene and their clear association with carcass parameters validates the gene's functional relevance in broiler genetic improvement. Birds possessing favourable *MSTN* alleles displayed superior carcass parameters. In AA genotype, birds with Adenine and Thymine alleles should be selected and reared for meat. In AAP and Cobb, only Adenine allele birds should be considered while birds possessing Adenine and Guanine alleles in Ross should be selected and raised for meat production. Therefore, Ross can be recommended having higher *MSTN* alleles which displayed superior carcass parameters confirmed suitability for high-yield meat production. Amino acid changes from proline to Arginine may possibly influence growth and skeletal muscle development in the population by enhancing nutrient delivery to muscles. Mutation changing from threonine to histidine may influence muscle growth, histidine is essential for growth, maintenance and repair of tissue. Because aspartic acid changing to arginine is a missense mutation which disrupts function of *Myostatin* leading to increased carcass weights, therefore Ross birds carrying this particular mutation should be used for selection.

Article Publication Details

This article is published in the **UAR Journal of Multidisciplinary Studies (UARJMS)**, ISSN 3049-4346 (Online). In Volume 1 (2025), Issue 10 (December)

The journal is published and managed by **UAR Publisher.**

Data Availability Statement

Raw data can be provided upon request.

Authors' Contributions

[1]Adewole R. A**.** is the corresponding author, she contributed to the research work in terms of conceptualization, designing of methodology, data collection and analysis, manuscript drafting, manuscript submission and publication process.

[2]Prof. Adedeji T. A. contributed in the supervision of the research work and review of the manuscript.

[3]Prof. Emiola I. A. contributed in the review and editing of the manuscript.

[4,5]Prof. Adebambo A. O. contributed in the data analysis of the research work.

Declarations

Ethical Approval

There was no breach of ethical or regulatory guidelines during the data collection of chicken carcasses and blood collection for molecular analysis, and the study posed no adverse impact on the community.

Funding Information

Authors declare that no funding was received for this research work.

Competing Interest

Authors declare that they have no competing interests.

References

Adebambo, A. O., Ikeobi, C. O. N., Ozoje, M. O., & Adebambo, O. A. (2010). Comparative study of growth performance and carcass characteristics of three commercial broiler strains in Nigeria. *Nigerian Journal of Animal Production*, *37*(1), 1–12.

Aviagen. (2021). Arbor Acre Plus: Top performance in the field. https://en.aviagen.com

Ayorinde, K. L., Oke, U. K., & Omoniyi, J. A. (2001). Methods of collecting poultry blood samples for research. *Animal Science Journal*, *20*(1), 55–59.

Bellinge, R. H. S., Liberles, D. A., Iaschi, S. P. A., O'Brien, P. A., & Tay, G. K. (2005). Myostatin and its implications on animal breeding: A review. *Journal of Animal Genetics*, *36*(1), 1–6.

Burt, D. W. (2005). Chicken genome: Current status and future opportunities. *Genome Research*, *15*(12), 1692–1698. https://doi.org/10.1101/gr.3973605

Fasiola, O. A., Adebambo, O. A., & Ikeobi, C. O. N. (2017). Genetic evaluation of growth and carcass traits in broiler chickens. *Tropical Animal Health and Production*, *49*(6), 1219–1225. https://doi.org/10.1007/s11250-017-1339-3

Fernanda, L., Castro, P., Sudhir, Y., Rebecca, L., Steven, C., Robert, P., & Woo, K. (2020). The effects of L-Arginine supplementation on growth performance and intestinal health of broiler chickens challenged with *Eimeria* spp. *Journal of Poultry Science*, *99*(11), 5844–5857. https://doi.org/10.3382/jps/pez602

Hakamata, Y., Toyomizu, M., & Kikusato, M. (2020). Differences in breast muscle mitochondrial respiratory capacity, reactive oxygen species generation, and complex characteristics between 7-week-old meat- and laying-type chickens. *The Journal of Poultry Science*, *57*(4), 319–327. https://doi.org/10.2141/jpsa.0190059

Iyakutye, J., & Hollinshead, H. (2021). Cysteine in broiler poultry nutrition. *Veterinary World*, *14*(8), 2244–2250. https://doi.org/10.14202/vetworld.2021.2244-2250

Khajali, F., & Wideman, R. F. (2010). Dietary arginine: Metabolic, environmental, immunological and physiological interrelationships. *World's Poultry Science Journal*, *66*(4), 751–766. https://doi.org/10.1017/S004393391000104X

Oke, O. E., Akosile, O. A., Uyanga, V. A., Oke, F. O., Oni, A. I., Tona, K., & Onagbesan, O. M. (2024). Climate change and broiler production. *Veterinary Medicine and Science*, *10*(3), Article e1416. https://doi.org/10.1002/vms3.1416

Okolo, G. P., Egena, S. S. A., Otu, B. O., & Sikiru, A. B. (2023). Expression of major histocompatibility complex class II gene in commercial strains of broiler chickens administered varying levels of aqueous ginger extract. *Nigerian Journal of Animal Production*, *49*(5), 147–155.

Oladokun, V. A., Adeyemo, G. O., & Hassan, O. A. (2020). Constraints to commercial poultry production in Nigeria. *Journal of Agricultural Economics and Development*, *9*(3), 55–62.

Olawunmi, O. O. (2020). Genetic and phenotypic assessment of broiler carcass traits. *African Journal of Genetics and Animal Breeding*, *8*(2), 115–123.

Ran, J. S., Lin, H. Y., & Wang, J. (2017). Role of Myostatin gene mutations in muscle growth of poultry. *Poultry Science Reports*, *96*(12), 4325–4334.

Rasmussen, K. L. (2016). *Investigation of variation in the promoter region of the myostatin gene (GDF-8) in New Zealand cattle* [Unpublished doctoral thesis, Lincoln University].

Ravi Kambadur, R. K., Bishop, A., Salerno, M. S., McCroskery, S. E. U. M. A. S., & Mridula, S. (2004). Role of Myostatin in muscle growth. In *Muscle development of livestock animals: Physiology, genetics and meat quality* (pp. 297–316). CABI Publishing.

Rubin, D. M., O'Reilly, A. L., Luan, X., & Localio, A. R. (2007). The impact of placement stability on behavioural well-being for children in foster care. *Journal of Pediatrics*, *119*(2), 336–344.

Sam, I. M., & Okon, L. S. (2022). Comparative evaluation of growth performance, morphometric and carcass traits of three strains of broiler chicken raised in the tropics. *Animal Research International*, *19*(3), 4617–4624.

Sartika, T., Suryani, S., & Purwati, E. (2011). Genetic polymorphisms of *MSTN* gene in indigenous and commercial chickens. *Indonesian Journal of Animal and Veterinary Sciences*, *16*(4), 253–261.

SAS Institute. (2023). Statistical Analysis System (SAS) Users guide (Version 9.4).

Sewalem, A., Morrice, D. M., Law, A., Windsor, D., Haley, C. S., Ikeobi, C. O., & Hocking, P. M. (2002). Mapping of quantitative trait loci for body weight at three, six, and nine weeks of age in a broiler layer cross. *Poultry Science*, *81*(12), 1775–1781. https://doi.org/10.1093/ps/81.12.1775

Sharma, P., Mishra, S. K., & Singh, Y. (2015). Advances in poultry genomics and marker-assisted selection. *Veterinary World*, *8*(5), 585–591. https://doi.org/10.14202/vetworld.2015.585-591

Tahmasbi, P. (2024). *Investigation in the relationship between competition, quality, price, market share, and financial efficiency in the poultry industry* [Doctoral thesis, University of Essex].

Tanjung, L. R., Sulandari, S., & Sartika, T. (2019). Primer design and PCR optimization for Myostatin gene amplification in chickens. *Indonesian Journal of Biotechnology*, *24*(1), 29–36. https://doi.org/10.22146/ijbiotech.41508

Zhang, S. Z., Penner, G. B., Yang, W. Z., & Oba, M. (2010). Effects of partially replacing barley silage or barley grain with dried distillers' grains with soluble on rumen fermentation and milk production of lactating dairy cows. *Journal of Dairy Science*, *93*(7), 3231–3242. https://doi.org/10.3168/jds.2009-2993

Zhang, Y., Xu, D., Dai, Y., & Zhang, J. (2018). Regulation of Myostatin expression is associated with muscle development in chickens. *Journal of Poultry Science*, *97*(6), 2230–2237. https://doi.org/10.3382/ps/pey186

Zhou, H., Mitchell, A. D., McMurtry, J. P., Ashwell, C. M., & Lamont, S. J. (2006). Association of Myostatin gene polymorphisms with growth traits in broilers. *Journal of Poultry Science*, *85*(3), 347–352. https://doi.org/10.1093/ps/85.3.347

Zerehdaran, S., Vereijken, A. L. J., van Arendonk, J. A. M., & Van der Waaijt, E. H. (2004). Estimation of genetic parameters for fat and protein efficiency in broilers. *Journal of Poultry Science*, *83*(4), 521–525. https://doi.org/10.1093/ps/83.4.521