

STEPWISE CANONICAL DISCRIMINANT ANALYSIS FOR MORPHOMETRIC CHARACTERIZATION OF THREE STRAINS OF BROILER CHICKEN

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Isaac U. C., N. J. Okafor, B. C. Nwachukwu, J. C. Albert, C.F. Aniemen, C. A. Igbokwe (2024). *Stepwise canonical discriminant analysis for morphometric characterization of three strains of broiler chicken*. - Genetika, Vol 56, No.1, 43-54.

Stepwise canonical discriminant analysis of quantitative traits was performed to characterize Arbor Acre (n = 29), Marshall (n = 29) and Ross 308 (n = 30) strains of broiler chicken at two weeks of age. Average body weight of 322.11 ± 51.33 g, 310.70 ± 51.20 g and 310.24 ± 42.37 g were obtained for Arbor Acre, Marshall and Ross 308 strains, respectively. Correlation coefficients between traits was positive (0.13-0.80). Out of the eight traits used, shank length (SL), body length (BL), wing length (WL), breast girth (BG) and thigh circumference (TC) were selected by stepwise procedure as discriminating variables with respective tolerance of 0.75, 0.84, 0.86, 0.57 and 0.66. The Mahalanobis distance indicated that Arbor Acre and Marshall were closely related by SL (2.006), WL (5.704), BG (6.002) and TC (6.314) while Arbor Acre and Ross 308 were related by BL (4.212). Two canonical discriminant functions were generated. Function 1, with smaller Wilk's Lambda (0.15), stronger canonical correlation coefficient (0.88), highly significant Chi-square ($p < 0.0001$), greater eigenvalue (3.14) and percentage variance (87.28%), possessed higher discriminatory power than function 2. 72.4%, 82.8% and 90.0% of the cross validated cases were correctly classified as Arbor Acre, Marshall and Ross broilers, respectively. The study revealed that Ross 308 was genetically distinct from Arbor Acre and Marshall, which were more closely related, using shank length, body length, wing length, breast girth and thigh circumference as discriminating traits. Stepwise canonical discriminant analysis is therefore relevant in classification genetics.

Key words: Broilers, quantitative traits, genetic relationship, classification

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INTRODUCTION

Poultry meat is the most consumed worldwide (SHAHBANDEH, 2023). The global population is projected to be about 9 billion by 2050, implying that an increase in income among the poor populace will lead to an unprecedented increase in animal protein demand. In this context, the poultry industry has an important role to play in sustainable food supply, and should be given prior consideration in improvement studies. Quantitative traits are good indicators of growth and market value of broilers (ISAAC *et al.*, 2023). They serve as basis for characterization, classification, selection and improvement of poultry breeds (AJAYI *et al.* 2012; N'DRI *et al.* 2016).

Discriminant analysis is an approach for analyzing performance data of broiler chickens. BARBOSA (2005) reported that the techniques of discriminant analysis have been successfully employed as a means of identifying developing genotypes and better utilize the advantages provided by heterosis. Canonical discriminant analysis is a multivariate technique that describes the relationship between two or more variables through linear combinations that are maximally correlated (OGAH, 2013). The goal of discriminant analysis is to elucidate how variation among groups is maximized and variation within group is minimized along a gradient, thus enhancing the knowledge of the genetic relatedness and diversity in poultry (AZIZ and AL-HUR, 2013; ZAITOUN *et al.*, 2005). The use of stepwise approach in multivariate discriminant analysis is essential for reduction of interdependency among a set of traits that are correlated (ISAAC and ADEOLU, 2023). Multivariate discriminant analysis of morphometric traits has been successfully used to estimate genetic variation within and between local breeds (KADURUMBA *et al.*, 2014; ZAITOUN *et al.*, 2005). ABDELQADER *et al.* (2010) reported that body weight, body length, heart girth and height at hip showed the largest discriminatory power between three Jordanian chicken genotypes.

YAKUBU *et al.* (2010) used discriminant analysis to correctly classify West African Dwarf and Red Sokoto goat populations of Nigeria into their source population. In today's poultry industry and breeding companies, there exist many strains of broilers that have compromised their genetic integrity, purity and performance. There is need to discriminate today's broiler strains into distinct genetic groups and at the same time group related strains together based on their common attributes. This will foster conservation of genetic resources and improvement. The objectives of the study were to: (i) evaluate the growth performance of Arbor Acre, Marshall and Ross 308 strains based on their quantitative traits, (ii) determine the best traits for discriminating among the three strains, (iii) correctly classify the broilers into different strains and (iv) determine the genetic relatedness or otherwise among the strains.

MATERIALS AND METHODS

Experimental location

The experiment was carried out at the Teaching and Research Farm of the Department of Animal Science and Technology, Faculty of Agriculture, Nnamdi Azikiwe University, Awka, Anambra State, Nigeria. The University is located within the tropical rainforest zone of south-eastern part of Nigeria on Latitude 6°24' N, Longitude 7°25' E and altitude 75 to 77 m. This area has average daily rainfall of 1.037 mm, annual rainfall of 1600 mm and minimum and maximum

daily ambient temperatures of 26.5°C and 34.7°C, respectively and average annual relative humidity of the area is 83%.

Procurement and management of experimental birds

Three commercial strains of day-old broiler chicks of Ross 308, Arbor Acre and Marshall were purchased from two renowned commercial farms in Nigeria, through their distributors and were transported at the cool hour of the night to minimize stress on the animals. The day-old chicks were a total of 90 in number, comprising 30 each of Arbor Acre, Marshall and Ross 308 strains.

The chicks were managed intensively in a deep litter, open sided, dwarf walled house, measuring 4.2 m x 2.2 m x 0.7 m. The chicks were fed commercial starter feed containing 22% crude protein and 2,900 kcal metabolizable energy/kg of feed. Feed and water were offered to the chickens *ad libitum*.

Data collection and measurements of traits

Data were taken at week two on individual broiler chicken on body weight (BW), body length (BL), Wing length (WL), wing span (WS), shank length (SL), breast girth (BG), keel length (KL) and thigh circumference (TC). The measurements of traits were described according to ISAAC *et al.* (2022).

Statistical analysis

Stepwise canonical discriminant analysis was carried out using IBM SPSS STATISTICS (2011) Computer Software. The general model of the linear discriminant function was given in expression (1).

$$Y = a + l_1 x_1 + l_2 x_2 + \dots + l_p \quad \dots (1)$$

where,

Y = Discriminant z score of discriminant function

a = Intercept

l = Discriminant coefficient for a quantitative trait

x = Any quantitative trait

The analysis provided the following statistics among others.

Tolerance

Tolerance is the proportion of the variance of a variable not accounted for by other independent variables in the equation (SPSS, 2006). It is used to determine how much the variables (traits) are linearly related to one another. In other words, it determines the level of multicollinearity existing among the observed traits. High tolerance indicates that a variable or trait is quite independent of other variables and contributes much information to a model (SPSS, 2006).

Tolerance was computed using expression (2)

$$1-R^2 \quad \dots (2)$$

where R^2 is the coefficient of determination, which is the percentage contribution of a particular quantitative trait to the total variation in performance (ISAAC and ADEOLU, 2023). R^2 was computed as given by YAKUBU *et al.* (2022).

Mahalanobis distance

This was calculated as a squared distance between any two strains in terms of a given trait studied. Mahalanobis distance was calculated according to MAHALANOBIS (1936) as specified in expression (3).

$$D^2 = (x_1 - x_2) \text{Cov}^{-1} (x_1 - x_2) \dots (3)$$

where,

D^2 = Mahalanobis distance (squared)

X_1 and X_2 = means of any trait of strain 1 and 2 respectively

Cov^{-1} = inverse of covariance of a trait in any two strains. Covariance was computed according to the formula given by ISAAC (2020).

Eigenvalue (variance ratio)

It was obtained as the ratio of the distance between strain sum of squares (SPSS, 2006).

Wilks' Lambda

It was computed as the proportion of the total variance in the discriminant scores not explained by differences among the groups (strains). The value ranges between 0 and 1. Values close to 0 indicate that group means are different.

RESULTS AND DISCUSSION

Means (\pm standard deviation)

Means and standard deviation of the quantitative traits of the three strains of broiler chickens are presented in Table 1. Arbor Acre recorded higher body weight (322.11 ± 51.33 g) and body length (19.26 ± 1.07 cm) than Marshall (310.51 ± 02 g; 18.21 ± 1.13 cm) and Ross 308 (310.24 ± 42.37 g; 18.21 ± 1.13 cm) in most other linear body parameters. This result agrees with previous reports (UDEH and OGBU, 2011; SAM and OKON, 2022).

Table 1. Mean (standard deviation) quantitative traits of the three broiler strains

Strain	Trait							
	BW (g)	BL (cm)	KL(cm)	BG(cm)	WL(cm)	WS (cm)	TC (cm)	SL (cm)
Arbor	322.11	19.26	8.02	16.21	10.89	24.21	6.89	3.27
Acre	(51.33)	(1.07)	(0.66)	(1.04)	(0.58)	(1.51)	(0.91)	(0.14)
Marshall	310.70	17.76	7.89	16.60	10.52	24.45	6.84	3.44
	(51.02)	(0.90)	(0.66)	(1.27)	(0.58)	(1.08)	(0.43)	(0.09)
Ross 308	310.24	18.21	7.68	16.18	11.24	25.18	7.15	3.05
	(42.37)	(1.13)	(0.81)	(0.86)	(0.67)	(0.86)	(0.45)	(0.11)

BW = Body weight, BL = Body length, KL = keel length, BG = Body girth, WL = Wing length WS = Wing span, TC = Thigh circumference, SL = Shank length

Correlation of traits pooled within strains

The correlation coefficients presented in Table 2 are pooled from the three strains as obtained from the discriminant analysis. Individual strain results are therefore, not presented. The traits all had positive correlation with each other. However, the strongest association

observed was between body weight and body girth ($r = 0.80$). This implies that body weight can greatly be improved by indirect selection of strains with larger breast girth. The result collaborates with the findings of OJEDAPO *et al.* (2012), and has informed the use of breast width as an important estimator of body weight and selection criterion for meat type birds (FAYEYE *et al.*, 2014; ISAAC, 2020; ISAAC and OBIKE, 2020). Other traits with high correlation with body weight were thigh circumference ($r = 0.66$), wing span ($r = 0.58$) and shank length ($r = 0.52$). These traits can be used in a selection index for improvement of high body weight on broilers (SADICK *et al.*, 2020). In addition, thigh circumference, wing span, body girth and wing length correlated positively among themselves. Improvement of any of these traits can positively affect others through correlated response.

Table 2. Correlation between traits pooled within strains

	BW	BL	KL	BG	WL	WS	TC	SL
BW	1.00							
BL	0.43	1.00						
KL	0.41	0.30	1.00					
BG	0.80	0.40	0.33	1.00				
WL	0.43	0.18	0.16	0.29	1.00			
WS	0.58	0.21	0.27	0.52	0.53	1.00		
TC	0.66	0.23	0.38	0.55	0.26	0.47	1.00	
SL	0.52	0.17	0.13	0.42	0.31	0.33	0.39	1.00

BW = Body weight, BL = Body length, KL = keel length, BG = Body girth, WL = Wing length WS = Wing span, TC = Thigh circumference, SL = Shank length

Mahalanobis distance between strains

Stepwise discriminant analysis revealed the traits that maximized the Mahalanobis distance (Table 3). From the analysis, Arbor Acre and Marshall were more closely related in shank length (2.006), wing length (5.704), body girth (6.002) and thigh circumference (6.314) while Arbor Acre and Ross 308 were closely related only in body length (4.212). This suggests more genetic similarity or relatedness between Arbor Acre and Marshall than between Ross 308. From the result, there seems to exist a common genetic base or origin for Arbor Acre and Marshall broiler chickens. It further underscores the usefulness of Mahalanobis distance for establishing genetic similarity or difference between breeds or strains (AZIZ and AL-HUR, 2013). LI *et al.* (2019) also noted that Mahalanobis distance is applied in data mining such as clustering and classification. Shank length which minimizes the distance between Arbor Acre and Marshall may serve as a reliable morphometric trait for classifying strains into same group among different sub-populations. This finding is supported by the research carried out by MUSHI *et al.* (2020) on Tanzanian free-range local chickens.

Table 3. Traits that maximize the Mahalanobis distance between two closest strains by stepwise discriminant analysis

Step	Trait	MDS	Between	Exact F statistics	df1	df2	Sig.
1	SL	2.006	Arbor Acre and Marshall	29.096	1	85.00	6.12 x10 ⁻⁷
2	BL	4.212	Arbor Acre and Ross 308	30.691	2	84.00	9.87 x10 ⁻¹¹
3	WL	5.704	Arbor Acre and Marshall	26.919	3	83.00	2.95 x10 ⁻¹²
4	BG	6.002	Arbor Acre and Marshall	20.988	4	82.00	6.09 x10 ⁻¹²
5	TC	6.314	Arbor Acre and Marshall	17.448	5	81.00	1.09 x10 ⁻¹¹

MDS = Minimum Distance squared

BW = Body weight, BL = Body length, KL = keel length, BG = Body girth, WL = Wing length WS = Wing span, TC = Thigh circumference, SL = Shank length, F = Fisher, df1, df2 = degrees of freedom 1 and 2, respectively, sig. = Significance

The best discriminating traits

Stepwise discriminant analysis selected five out of the eight traits as the best discriminating traits and they are shank length (SL), body length (BL), wing length (WL), breast girth (BG) and thigh circumference (TC). These traits were used as discriminatory variables for the strains studied. Their tolerance values are presented in Table 4. The result underscores the importance of stepwise in multivariate discriminant analysis. In essence, it helps to exclude unwanted variables that are capable of increasing multicollinearity among a set of independent variables. Multicollinearity or interdependency reduces the accuracy of analysis and may lead to wrong inference on variables which did not contribute much to the overall variation in the performance of animals (KIM, 2019; ISAAC *et al.*, 2022).

Table 4. Traits selected by stepwise discriminant analysis for classifying strains into closely related groups

Step	Tolerance	F to Remove	MDS	Between Groups
1	SL	1.00	83.44	
2	SL	0.97	86.11	0.19
	BL	0.97	17.51	2.01
3	SL	0.89	91.86	1.41
	BL	0.95	15.77	3.21
	WL	0.89	11.95	4.21
4	SL	0.78	82.18	1.65
	BL	0.84	19.64	3.21
	WL	0.87	9.98	5.10
	BG	0.69	4.62	5.70
5	SL	0.75	84.60	1.86
	BL	0.84	19.40	3.53
	WL	0.86	7.69	5.50
	BG	0.57	2.77	5.78
	TC	0.66	3.92	6.00

MDS = Minimum Distance squared, BW = Body weight, BL = Body length, KL = keel length, BG = Body girth, WL = Wing length WS = Wing span, TC = Thigh circumference, SL = Shank length, F = Fisher

The best discriminating traits (body length, breast girth and shank length) in the present study have earlier been confirmed (ABDELQADER *et al.*, 2008). In a related study, AJAYI *et al.* (2012) also reported that breast girth, keel length, shank length and wing length were among some of the discriminating variables used in classifying chickens into distinct populations. However, OGAH (2013) reported body weight, thigh length and body width as the most discriminant variables for discriminating between normal feathered, frizzled and naked neck chickens. Differences in size and method of discriminant analysis employed by this author could account for this difference.

The high tolerance value (1.00) of shank length in step 1 (Table 4) indicated that it contributed the highest discriminatory power and variability independent of other traits. This explains the reason shank length has been recommended for phenotypic characterization of indigenous chickens in a previous study (MAHARANI *et al.*, 2021).

Canonical discriminant functions

Function 1 in Table 5 indicated that the strains were genetically different since its Wilks' Lambda (0.15) was closer to zero and smaller than 0.6 of function 2. This was highly significant ($p < 0.0001$) as evidenced by Chi-square (Bartlett's) test. The significance of Wilks' Lambda validated the discriminant analysis carried out. This is similar to what OGAH *et al.* (2011) reported.

Table 5. Summary of canonical discriminant function analysis

Function	Eigenvalue	% of Variance	Canonical Correlation	Test of Function(s)	Wilks' Lambda	Chi-square	df	Sig.
1	3.41	87.28	0.88	1 through 2	0.15	156.62	10	0.000
2	0.50	12.72	0.58	2	0.67	33.48	4	0.000

F = Fisher, Sig. = Significance

The provision of functions 1 and 2 in the summary of the canonical discriminant function analysis is actually due to three strains used, as the number of functions is usually one less than the number of discriminating groups. This agrees with the findings of OGAH (2013). Function 1 had better discriminatory power judging from its higher eigenvalue or variance ratio (3.14) and percentage variance (87.28%) compared to function 2 with lower eigenvalue (0.50) and percentage variance (12.72%) variance. This result also concurs with the research conducted by ARIZA *et al.* (2022) who reported that lower values of Wilks' lambda indicated a better discriminatory power.

Functions at group centroid

The functions at group centroid are presented in Table 6. The centroid was considered as the group means of the predictor variables or morphometric traits of chickens (cases) in each strain. Chickens with scores near to a centroid are regarded as belonging to one group (OGBOGO, 2019). Group centroids are therefore important statistics for accurate classification of individuals.

Table 6. Functions at group centroid

Strain	Function	
	1	2
Arbor Acre	0.17	0.99
Marshall	2.17	-0.54
Ross 308	-2.26	-0.43

The positive signs for Arbor Acre and Marshall and the negative for Ross 308 in Function 1 indicated that Arbor Acre and Marshall are in one direction and opposite to Ross 308, suggesting that Arbor Acre and Marshall were closely related but differed greatly from Ross 308 based on the morphometric traits measured. Arbor Acre and Marshall can therefore be classified into the same genetic group. The magnitude of the values in function 1 indicated that the mean morphometric traits of Marshall broilers (2.17) was higher than that of Arbor Acre (0.17).

Classification function coefficients

The coefficients of the function are shown in the Table 7. From the table, the best reduced discriminant models for discriminating among the strains are as follows.

Arbor Acre: $D = -528.16 + 13.71BL - 1.51BG + 14.26WL - 5.61TC + 213.26SL$

Marshall: $D = -540.47 + 11.95BL - 0.60BG + 12.67WL - 6.69TC + 229.45SL$

Ross 308: $D = -473.09 + 12.47BL - 0.84BG + 16.20WL - 3.85TC + 189.03SL$

The coefficients indicated the contributions of each morphometric trait to the discriminant function (equation). Shank length, which had the highest contribution in each strain, is regarded as the best trait for strain separation or discrimination.

Table 7. Classification function coefficients

Trait	Strain		
	Arbor Acre	Marshall	Ross 308
BL	13.71	11.95	12.47
BG	-1.51	-0.60	-0.84
WL	14.26	12.67	16.20
TC	-5.61	-6.69	-3.85
SL	213.26	229.45	189.03
(Constant)	-528.16	-540.47	-473.09

Classification results

The classification results (Table 8) revealed that 23 (79.3%), 25 (86.2%) and 27 (90.0%) out of 29, 29 and 30 original cases were correctly classified as Arbor Acre, Marshall and Ross 308 strains respectively. This means that out of the original cases counted, a total of 6 Arbor Acre were wrongly classified, 3 (10.3%) each as Marshall and Ross 308, 3 (10.3%) Marshall and 1 (3.4%) Marshall were wrongly classified as Arbor Acre and Ross 308, respectively, 2 (6.7%) Ross 308 and 1 (3.3%) Ross 308 were wrongly classified as Arbor Acre and Marshall strains respectively. However, cross validated cases, which provide more honest presentation of the

discriminant function, indicated that 21 (72.4%), 24 (82.8%) and 27 (90.0%) cases were correctly classified as Arbor Acre, Marshall and Ross 308 strains.

Table 8. Classification results

				Predicted group membership			Total
Strain			Arbor Acre	Marshall	Ross 308		
Cases Selected	Original	Count	Arbor Acre	23	3	3	29
			Marshall	3	25	1	29
			Ross 308	2	1	27	30
	Percentage	Arbor Acre	79.3	10.3	10.3	100	
		Marshall	10.3	86.2	3.4	100	
		Ross 308	6.7	3.3	90.0	100	
Cross validated	Count	Arbor Acre	21	4	4	29	
		Marshall	4	24	1	29	
		Ross 308	2	1	27	30	
	Percentage	Arbor Acre	72.4	13.8	13.8	100	
		Marshall	13.8	82.8	3.4	100	
		Ross 308	6.7	3.3	90.0	100	
Cases not selected	Original	Count	Arbor Acre	6	59	0	65
			Marshall	0	74	1	75
			Ross 308	1	83	0	84
	Percentage	Arbor Acre	9.2	90.8	0.0	100	
		Marshall	0.0	98.7	1.3	100	
		Ross 308	1.2	98.8	0.0	100	

Ross 308 individuals, thus had the greatest predictive accuracy compared to the other two strains. Ross 308 broiler chickens were more unique in their quantitative characters than Arbor Acre and Marshall strains. The implication is that Ross broilers can easily be identified in a mixed population and classified with high fidelity based on their morphometric traits. Classification result is an essential tool for discriminating animals with doubtful origin or in mixed population. This is important for effective management and conservation (KADURUMBA *et al.*, 2014). Discriminant function classification has been used to characterize West African Dwarf and Red Sokoto goats in Nigeria (YAKUBU *et al.*, 2010) and village goat production systems in South Africa (MDLADLA *et al.*, 2017).

CONCLUSION

Discriminant analysis is suitable for evaluation of broiler chicken performance. The use of discriminant analysis in evaluating quantitative traits among the three broiler strains has helped in understanding their genetic relatedness and diversity. Of the eight quantitative traits used, five stood out as the best discriminatory traits which are body length, shank length, wing length, breast girth and thigh circumference. Arbor Acre and Marshall strains had more traits in common and were closely related while Ross 308 strain was quite different from the former two. This

result suggests that Arbor Acre and Marshall may have evolved or originated from a common genetic base.

ACKNOWLEDGEMENT

The authors are grateful to the Department of Animal Science and Technology for providing a conducive environment for this study.

Received, November 05th, 2023

Accepted, April 28th, 2024

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**STEPWISE KANONIČKA DISKRIMINANTNA ANALIZA ZA MORFOMETRIJSKU
KARAKTERIZACIJU TRI SOJA PILIĆA BROJLERA**

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Izvod

Urađena je postepena kanonska diskriminantna analiza kvantitativnih osobina da bi se okarakterisali Arbor Acre (n = 29), Marshall (n = 29) i Ross 308 (n = 30) sojevi brojlera u dobi od dve nedelje. Prosečna telesna težina od $322,11 \pm 51,33$ g, $310,70 \pm 51,20$ g i $310,24 \pm 42,37$ g dobijena je za sojeve Arbor Acre, Marshall i Ross 308, respektivno. Koeficijenti korelacije između osobina su bili pozitivni (0,13-0,80). Od osam korišćenih osobina, dužina trupa (SL), dužina tela (BL), dužina krila (VL), obim grudi (BG) i obim butina (TC) odabrani su postupnom procedurom kao diskriminatorne varijable sa odgovarajućom tolerancijom od 0,75, 0,84, 0,86, 0,57 i 0,66. Mahalanobisovo rastojanje je pokazalo da su Arbor Acre i Maršal blisko povezani sa SL (2.006), VL (5.704), BG (6.002) i TC (6.314), dok su Arbor Acre i Ros 308 povezani sa BL (4.212). Generisane su dve kanonske diskriminantne funkcije. Funkcija 1, sa manjom Vilkovom lambda (0,15), jačim koeficijentom kanonske korelacije (0,88), visoko značajnim hi-kvadratom ($p < 0,0001$), većom sopstvenom vrednošću (3,14) i procentualno varijansom (87,28%), posedovala je veću diskriminatornu moć od funkcije 2, 722,7. 4%, 82,8% i 90,0% ukrštenih validiranih slučajeva ispravno je klasifikovano kao brojleri Arbor Acre, Marshall i Ross, respektivno. Studija je otkrila da se Ros 308 genetski razlikovao od Arbor Acre i Maršala, koji su bili bliže povezani, koristeći dužinu trupa, dužinu tela, dužinu krila, obim grudi i obim butina kao diskriminatorne osobine. Postepena kanonska diskriminantna analiza je stoga relevantna u klasifikacionoj genetici.

Primljeno 05.XI.2023.

Odobreno 28. IV 2024.