



Egg-based typology of local chickens in Kongo Central, Democratic Republic of the Congo: morphometric traits and internal quality assessed using Haugh units

[Typologie des poules locales du Kongo Central, République démocratique du Congo, basée sur les œufs : caractéristiques morphométriques et qualité interne évaluées à l'aide des unités Haugh]

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Abstract

Laying performance and egg quality of local chickens remain poorly documented in the Democratic Republic of the Congo despite their importance as animal genetic and socioeconomic resources. To characterize eggs from local hens in Mbanza-Ngungu, Muanda, and Songololo and to establish an exploratory hen typology based on egg morphometric traits and Haugh units. A total of 190 eggs from 38 hens (five eggs per hen) were analyzed. Egg weight, length, width, and thick albumen height were measured; shape index and Haugh units were calculated. Descriptive analyses were performed at the egg level, whereas the main territorial comparisons, principal component analysis, and hierarchical clustering used hen-level means to account for repeated eggs from the same hen. Mean egg weight ranged from 34.13 ± 7.02 g in Songololo to 36.21 ± 6.60 g in Mbanza-Ngungu, with no robust territorial difference at the hen level. Haugh units differed among territories and were highest in Muanda. The first two principal components explained 79.60% of total variance. Two exploratory hen profiles were identified, differentiated mainly by egg size and, secondarily, by Haugh units; cluster separation was moderate and cluster membership was not associated with territory. The observed phenotypic variability provides a preliminary basis for individual-level characterization of local hens. Although eligibility criteria and the general feeding system were described, detailed individual covariates and post-laying conditions were insufficiently documented for adjustment. The typology remains exploratory and does not establish genetic differentiation or justify selection decisions without further validation.

Keywords : extensive poultry systems, egg freshness, phenotypic diversity, animal genetic resources, local selection

Résumé

En République démocratique du Congo, les performances de ponte et la qualité des œufs des poules locales restent mal documentées, malgré leur importance en tant que ressources génétiques animales et socio-économiques. Cette étude visait à caractériser les œufs de poules locales de Mbanza-Ngungu, Muanda et Songololo et à établir une typologie exploratoire des poules basée sur les caractéristiques morphométriques des œufs et l'indice de Haugh. Au total, 190 œufs provenant de 38 poules (cinq œufs par poule) ont été analysés. Le poids, la longueur, la largeur et la hauteur de l'albumen épais ont été mesurés ; l'indice de forme et l'indice de Haugh ont été calculés. Les analyses descriptives ont été réalisées au niveau de l'œuf, tandis que les principales comparaisons territoriales, l'analyse en composantes principales et la classification hiérarchique ont utilisé les moyennes au niveau de la poule afin de tenir compte des œufs répétés d'une même poule. Le poids moyen des œufs variait de $34,13 \pm 7,02$ g à Songololo à $36,21 \pm 6,60$ g à Mbanza-Ngungu, sans différence territoriale significative au niveau de la poule. Les unités Haugh variaient selon les territoires et étaient les plus élevées à Muanda. Les deux premières composantes principales expliquaient 79,60 % de la variance totale. Deux profils exploratoires de poules ont été identifiés, différenciés principalement par la taille des œufs et, secondairement, par les unités Haugh ; la séparation des groupes était modérée et l'appartenance à un groupe n'était pas associée au territoire. La variabilité phénotypique observée fournit une base préliminaire pour la caractérisation individuelle des poules locales. Bien que les critères d'éligibilité et le système d'alimentation général aient été décrits, les covariables individuelles détaillées et les conditions post-ponte n'étaient pas suffisamment documentées pour permettre un ajustement. La typologie demeure exploratoire et n'établit pas de différenciation génétique ni ne justifie les décisions de sélection sans validation supplémentaire.

Mots-clés : systèmes avicoles extensifs ; fraîcheur des œufs ; diversité phénotypique ; ressources génétiques animales ; sélection locale..

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1. Introduction

Family poultry farming plays a central role in low-input production systems in sub-Saharan Africa. Local chickens simultaneously fulfill nutritional, economic, social, and cultural functions. They provide accessible animal protein, constitute readily available household savings, and contribute to resilience against economic shocks. Their hardiness, ability to use locally available feed resources, and adaptation to challenging environments explain their persistence on family farms despite generally lower performance than specialized commercial strains (Guèye, 2000; Wong et al., 2017).

In the Democratic Republic of Congo, particularly in Kongo Central, local chickens are predominantly reared under extensive or semi-extensive systems. These systems are characterized by free ranging, irregular feed supplementation, limited health management, and poor control of reproduction. Nevertheless, local populations may harbor phenotypic diversity that is useful for gradual production improvement, provided that their traits are measured under their actual production conditions and interpreted cautiously. Egg characterization therefore provides a complementary approach to the morphological description of birds.

Egg quality is multidimensional. External traits, including weight, length, width, and shape index, influence marketability, mechanical resistance, incubation, and chick size. Internal traits provide information on yolk integrity and albumen quality. The Haugh unit, proposed by Haugh (1937), combines thick albumen height and egg weight and remains one of the most widely used indicators of internal egg quality and freshness (Jones, 2015). A high value generally reflects firmer albumen; however, it must be interpreted in relation to hen age, genotype, temperature, the interval between laying and measurement, and storage conditions (Williams, 1992; Silversides & Scott, 2001). Egg size and albumen measurements capture only part of egg quality; shell integrity, yolk traits, and handling conditions also matter (Roberts, 2004).

African studies illustrate variation in egg quality under different production conditions. In Morogoro, Tanzania, Nonga et al. (2010) reported an overall mean egg weight of 40.8 g, an albumen height of 3.92 mm, and a Haugh unit of 67.7 in a sample combining eggs from households and market sellers. In Kabwe, Zambia, Liswaniso et al. (2020) reported heavier eggs from indigenous free-range chickens and used PCA to summarize relationships among egg traits. Tadesse et

al. (2015) compared internal and external egg quality across production systems in Ethiopia. Such studies provide useful benchmarks, but differences in sampling, bird populations, husbandry, and egg handling restrict direct comparison with Kongo Central.

Most characterizations rely on univariate egg-by-egg comparisons. However, multiple eggs from the same hen are not independent observations. Ignoring this structure may underestimate uncertainty and generate spurious statistical significance. Moreover, examining traits separately does not capture their joint organization. An approach based on hen-level means, combining principal component analysis and clustering, can identify homogeneous hen profiles without confounding within-hen repetition with between-hen variation. Accounting for the biological source of repeated observations is essential to avoid pseudoreplication (Hurlbert, 1984).

This study aimed to establish an exploratory typology of local hens from three territories of Kongo Central using egg morphometric traits and albumen quality assessed by Haugh units. Specifically, it sought to: (i) describe variability in egg weight, dimensions, albumen height, and Haugh units; (ii) compare these traits among Mbanza-Ngungu, Muanda, and Songololo; (iii) analyze their multivariate relationships; and (iv) identify typological groups of hens. The working hypothesis was that the joint variation in these traits could reveal statistical profiles that would not necessarily coincide with territories of origin.

2. Material and methods

2.1. Study area and period

The study was conducted in Kongo Central Province, Democratic Republic of the Congo, specifically in the territories of Mbanza-Ngungu, Muanda, and Songololo. These territories have contrasting agroecological conditions: Muanda is a tropical coastal zone, Songololo is subhumid, and Mbanza-Ngungu is a subhumid highland zone. Eggs were reported as collected from the second half of April to the end of August 2024. This calendar interval is not consistent with the stated duration of 90 days; the exact collection dates and duration therefore require verification against the original field records.

2.2. Study population and sampling framework

The study included local laying hens (*Gallus gallus domesticus*) maintained by households in Mbanza-Ngungu, Muanda, and Songololo, Kongo

Central Province, Democratic Republic of the Congo. Eligible hens were older than 8 months, apparently healthy, and had live body weights ranging from 900 to 1,450 g. They were maintained under farmers' usual husbandry practices and obtained feed by free-range scavenging, including household by-products and other locally available feed resources. The onset of laying occurred between 6 and 8 months of age. Only hens that had completed at least two laying cycles were retained for egg collection; this criterion described their laying history rather than a standardized position within the current laying cycle.

The sampling structure comprised three levels: territories as geographical strata, hens as clusters of repeated observations, and eggs as measurement units. All three study territories were included to ensure geographical coverage. The sample comprised 38 hens: 12 from Mbanza-Ngungu, 14 from Muanda, and 12 from Songololo. Five consecutive eggs were collected per hen, yielding 60, 70, and 60 eggs, respectively, for a total of 190 eggs.

Households were selected from local veterinary registries in the three study territories. Eligible households maintained at least five actively laying hens under extensive or semi-intensive production systems. Egg collection was restricted to households that had consented to participate. Participation was voluntary, and farmers provided free and informed consent, verbally or in writing, before data collection.

One hen was selected from each of the 38 participating households. Individual hens were identified using visible morphological features, and each egg was labelled by territory, household, hen, and egg number to ensure traceability. The procedures used to select households from the registries and hens within households require further documentation before selection can be described as random.

The sample size of 38 hens was determined by available resources and collection feasibility, with 12–14 hens per territory. This pragmatic sample was intended for exploratory phenotypic characterization; no a priori power calculation was documented.

Collecting five eggs per hen ensured equal contributions to the dataset but did not eliminate dependence among eggs from the same bird. Eggs were therefore treated as repeated measurement units, whereas hens were the primary statistical units. The principal territorial comparisons and typology development were based on hen-level means.

Including one hen per household avoided sampling multiple birds sharing the same farm environment. However, hen and household effects could not be separated, and the design did not allow a genetic component of the observed variance to be isolated.

2.3. Measurement of egg traits

Each egg was identified by territory of origin, hen number, and individual egg number. Weight was recorded in grams, length and width in centimeters, and thick albumen height in millimeters. Morphometric measurements were obtained using a scale and a caliper. Albumen height was measured after breaking the egg onto a flat surface, within the thick albumen between the yolk and the outer albumen margin (Jones, 2015; Tadesse et al., 2015). Exact instrument models, precision, and the interval between laying and measurement were unavailable in the analyzed dataset and were therefore considered methodological limitations when interpreting the findings.

Two composite indicators were then calculated: shape index, defined as the ratio of egg width to egg length, and the Haugh unit, used as an indicator of internal egg quality. Shape index (%) = (egg width/egg length) × 100.

$HU = 100 \times \log_{10} [H + 7.57 - 1.7 \times W^{0.37}]$, where HU is the Haugh unit, H is thick albumen height in millimeters, and W is egg weight in grams. The constant 7.57 corresponds to the form of the equation used in the analysis.

2.4. Statistical analyses

Before analysis, data completeness, value ranges, and identifier consistency were checked. Descriptive statistics were generated by territory, including sample sizes, means, standard deviations, medians, minima, and maxima. Egg-level comparisons were treated as exploratory because the five eggs contributed by each hen were not independent observations. The main territorial inference was therefore based on means calculated for each of the 38 hens. Depending on distributional characteristics and variance assumptions, one-way analysis of variance or the Kruskal–Wallis test was used; nonparametric post hoc comparisons were adjusted using Holm's method (Holm, 1979). Statistical significance was set at 5%.

Principal component analysis (Abdi & Williams, 2010; Jolliffe & Cadima, 2016) was performed at the hen level using four active variables: mean egg weight, mean length, mean width, and mean Haugh unit.

Variables were centered and standardized before analysis. Shape index was excluded from the active variables because it is mathematically derived from length and width. Albumen height was not entered jointly with the Haugh unit to limit redundancy. Agglomerative hierarchical clustering based on Euclidean distance and Ward's method (Ward, 1963) was then applied to the principal-component scores.

The number of clusters was selected by comparing silhouette coefficients for solutions ranging from two to six clusters (Rousseeuw, 1987). The association between typological group and territory was examined using the χ^2 test. Analyses were performed in Python using standard statistical processing and scientific visualization libraries.

2.5. Ethical considerations

The study involved egg collection from family poultry farms without a reported experimental intervention on the hens. Farmers were informed of the study objectives and provided free and informed consent, verbally or in writing, before data collection. The manuscript does not provide a formal ethics-committee approval or exemption reference or the specific local authorization obtained. Any applicable approval, exemption, or authorization should be verified against the original study records and reported according to the requirements of the target journal and competent institutions; no approval number should be inferred in its absence.

3. Results

3.1. Sample structure and descriptive characteristics

The 190 eggs analyzed were distributed among Mbanza-Ngungu (60 eggs), Muanda (70 eggs), and Songololo (60 eggs). Each hen contributed five eggs, providing an equal number of repeated egg measurements per hen, although the numbers of hens differed slightly among territories (12, 14, and 12, respectively). Egg weight ranged from 18.17 to 54.84 g across the entire sample. Mean values were similar among territories, with the highest mean weight in Mbanza-Ngungu and the lowest in Songololo (table 1).

Table 1. Egg characteristics by territory (mean $\pm$ standard deviation)

Trait	Mbanza-Ngungu (n = 60)	Muanda (n = 70)	Songololo (n = 60)
Weight (g)	36.21 $\pm$ 6.60	35.01 $\pm$ 7.58	34.13 $\pm$ 7.02
Length (cm)	4.99 $\pm$ 0.48	5.01 $\pm$ 0.44	5.17 $\pm$ 0.42
Width (cm)	3.36 $\pm$ 0.39	3.36 $\pm$ 0.35	3.52 $\pm$ 0.35
Shape index (%)	67.49 $\pm$ 5.95	67.18 $\pm$ 5.82	68.26 $\pm$ 5.86
Albumen height (mm)	4.59 $\pm$ 0.82 ¹	5.01 $\pm$ 0.53 ¹	4.57 $\pm$ 0.81 ¹
Haugh unit	75.69 $\pm$ 6.07 ¹	79.58 $\pm$ 4.33 ¹	76.77 $\pm$ 4.72 ¹

Note: Descriptive values in the table were calculated at the egg level (n = 60–70 per territory). However, the main inferential tests and conclusions were based on hen-level means (n = 12–14) to limit pseudoreplication. The superscript ¹ identifies variables for which the principal inferential results are discussed at the hen level.

3.2. Comparisons among territories

Egg weight and shape index did not differ significantly among the three territories in the reported analyses. At the egg level, length (ANOVA: p = 0.0417; Kruskal–Wallis: p = 0.0219) and width (ANOVA: p = 0.0148; Kruskal–Wallis: p = 0.0024) showed apparent territorial differences. These differences were not retained when the analysis used the 38 hen-level means (p = 0.2997 for length and p = 0.1815 for width). The egg-level p-values should therefore be interpreted as exploratory because treating repeated eggs from the same hen as independent observations violates the independence assumption.

Internal quality showed a different pattern. Mean hen-level Haugh units differed among territories by both ANOVA (p = 0.0121) and the Kruskal–Wallis test (p = 0.0043). Muanda had the highest mean (79.58 $\pm$ 2.64 at the hen level), followed by Songololo (76.77 $\pm$ 2.70) and Mbanza-Ngungu (75.69 $\pm$ 4.26). After Holm adjustment, Muanda differed from Mbanza-Ngungu (p = 0.0152) and Songololo (p = 0.0218), whereas Mbanza-Ngungu and Songololo did not differ (p = 0.2145). For albumen height, the Kruskal–Wallis test indicated a territorial difference at the hen level (p = 0.0282); the Mbanza-Ngungu–Muanda comparison remained significant after adjustment (p = 0.0281).

Table II. Summary of territorial comparisons.

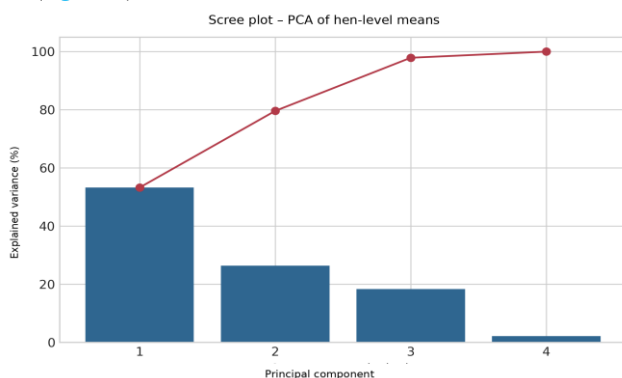
Variable	Egg-level test	Hen-level test	Conclusion
Weight	ANOVA $p = 0.2744$; KW $p = 0.6789$	ANOVA $p = 0.6184$	No robust difference
Length	ANOVA $p = 0.0417$; KW $p = 0.0219$	ANOVA $p = 0.2997$	Signal not confirmed
Width	ANOVA $p = 0.0148$; KW $p = 0.0024$	ANOVA $p = 0.1815$	Signal not confirmed
Shape index	ANOVA $p = 0.5668$; KW $p = 0.2567$	ANOVA $p = 0.4369$	No robust difference
Albumen height	KW $p = 0.00014$	KW $p = 0.0282$	Territorial difference at hen level
Haugh unit	ANOVA $p < 0.001$; KW $p < 0.001$	ANOVA $p = 0.0121$; KW $p = 0.0043$	Highest mean in Muanda

3.3. Relationships among egg traits

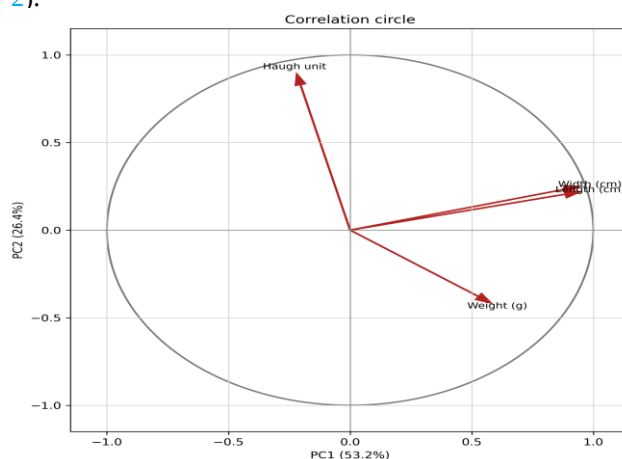
In the exploratory egg-level analysis ($n = 190$), length and width were positively correlated ($r = 0.626$). Weight showed positive but moderate correlations with length ($r = 0.296$) and width ($r = 0.300$). Shape index was positively associated with width ($r = 0.600$) and negatively associated with length ($r = -0.245$), consistent with its mathematical definition. Albumen height was strongly correlated with the Haugh unit ($r = 0.786$), whereas the Haugh unit was negatively correlated with egg weight ($r = -0.306$), which is partly expected because egg weight enters the Haugh-unit equation. Because these correlations were calculated from repeated eggs within hens, they should be interpreted descriptively rather than as independent-observation inferential estimates.

3.4. Factor structure

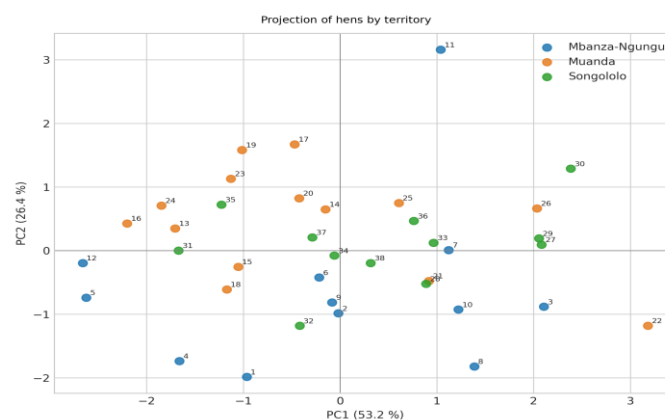
The PCA based on means from the 38 hens yielded a first component explaining 53.25% of variance and a second component explaining 26.35%. Together, they accounted for 79.60% of total variance. The third component explained an additional 18.29%, bringing cumulative variance for the first three components to 97.89%. The first two components were retained for the principal graphical interpretation (figure 1).

**Figure 1.** Scree plot from the PCA applied to hen-level means.

The first component was strongly and positively associated with length (0.950) and width (0.948), and more moderately with weight (0.582). It therefore represented a general egg-size gradient. The second component was primarily determined by Haugh units (0.895), whereas egg weight contributed negatively (-0.418). It thus contrasted hens producing eggs with better albumen quality against hens characterized by heavier eggs but relatively lower Haugh units (figure 2).

**Figure 2.** Correlation circle of active traits on the first two principal components.

The projection of hens by territory showed substantial overlap among the three origins, with no clear territorial separation on the factor map. Some hens nevertheless occupied relatively extreme positions on the retained components. This pattern is consistent with appreciable within-territory variability, but the PCA alone does not provide a formal test that within-territory variability exceeds between-territory variability (figure 3).

**Figure 3.** Projection of the 38 hens on the PC1–PC2 plane by territory.

3.5. Hen typology

Comparison of solutions containing two to six clusters selected the two-cluster solution, which had the highest silhouette coefficient. However, the silhouette value remained moderate (0.284), indicating imperfect separation and a continuum between profiles rather than fully distinct categories.

The first group comprised 27 hens producing smaller eggs, with a mean weight of 33.16 ± 4.06 g, length of 4.92 ± 0.27 cm, and width of 3.31 ± 0.21 cm. Their mean Haugh unit was 78.14 ± 3.80 . The second group comprised 11 hens producing larger and heavier eggs: 39.90 ± 4.31 g, 5.39 ± 0.17 cm long, and 3.66 ± 0.16 cm wide. Their Haugh unit was slightly lower at 75.82 ± 2.45 . The contrast therefore combined an egg-size gradient with a secondary albumen-quality gradient.

Table III. Profiles of the two typological groups of hens

Trait	Group 1 (n = 27)	Group 2 (n = 11)	Interpretation
Mean weight (g)	33.16 ± 4.06	39.90 ± 4.31	Heavier eggs in Group 2
Mean length (cm)	4.92 ± 0.27	5.39 ± 0.17	Longer eggs in Group 2
Mean width (cm)	3.31 ± 0.21	3.66 ± 0.16	Wider eggs in Group 2
Shape index (%)	67.45 ± 2.05	68.04 ± 2.41	Similar shape profiles
Albumen height (mm)	4.70 ± 0.62	4.83 ± 0.36	Limited difference
Haugh unit	78.14 ± 3.80	75.82 ± 2.45	Slightly higher in Group 1

Both groups occurred in each territory. Group 1 included eight hens from Mbanza-Ngungu, 11 from Muanda, and eight from Songololo; Group 2 included four, three, and four hens, respectively. The association between territory and group was not significant ($\chi^2 = 0.609$; $p = 0.737$). Thus, the typology did not coincide with geographical origin (figure 4).

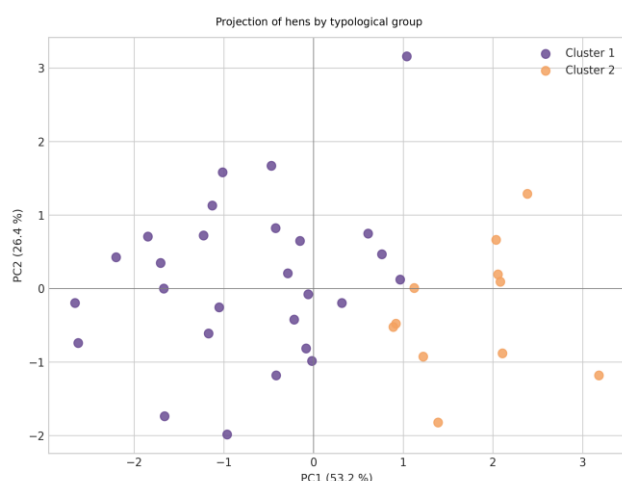


Figure 4. Projection of hens on the factor plane by typological group

The factorial projection of the two groups shows that the classification was driven mainly by the egg-

size gradient represented by the first principal component. Group 2 had higher scores on this component, corresponding to hens producing longer, wider, and heavier eggs, whereas Group 1 comprised mainly hens producing smaller eggs with slightly higher Haugh units. Separation remained partial, consistent with the silhouette coefficient of 0.284 and with an exploratory phenotypic continuum rather than sharply distinct categories. The classification therefore supports individual-level characterization using combined egg-size and albumen-quality criteria, but it does not establish biological breeds or genetic groups.

4. Discussion

4.1. Egg weight and morphometry

Mean egg weights, ranging from 34.13 to 36.21 g among territories, were lower than the overall mean of 40.8 g reported in the combined household-and-market sample from Morogoro, Tanzania (Nonga et al., 2010), and the 48.63 g reported in Kabwe, Zambia (Liswaniso et al., 2020). These are contextual comparisons rather than evidence of an intrinsic population difference: sampling and egg handling differed across studies.

Variation in hen age, body size, feed availability, or laying history may contribute, but the present study cannot isolate their effects (Roberts, 2004; Silversides & Scott, 2001).

The analysis did not detect a robust territorial difference in egg weight; this does not establish equivalence between territories. Results for length and width illustrate the issue of pseudoreplication: significant differences appeared when 190 eggs were treated as independent but disappeared when the 38 hen-level means were analyzed. Repeated eggs should be accounted for through hen-level summaries or a model that explicitly represents within-hen dependence (Hurlbert, 1984). A mixed-effects model including hen as a random effect and territory as a fixed effect would be an appropriate extension if data from more birds, laying dates, and covariates were available.

The positive correlation between length and width reflects a common dimensional factor. Weight was only moderately associated with these dimensions, suggesting that part of its variation also depends on internal composition and shell characteristics. Studies of indigenous Zambian chickens have generally reported stronger positive relationships between weight and several external and internal traits (Liswaniso et al., 2020). This difference may arise from the limited number of traits measured here and the substantial

within-hen heterogeneity observed in some five-egg series.

4.2. Albumen height and Haugh units

Mean Haugh units in all three territories exceeded 75 and were higher than the mean of 67.7 reported by Nonga et al. (2010) for their combined household-and-market sample in Tanzania. In the present sample, Muanda had the highest mean Haugh unit, reflecting the combination of greater albumen height and relatively moderate egg weight. These values indicate comparatively firm albumen at the time of measurement, but they should not be assigned a universal quality grade because laying-to-measurement interval and storage conditions were not documented.

The territorial difference in Haugh units must nevertheless be interpreted cautiously. This indicator is strongly influenced by the interval between laying and analysis, storage temperature, hen age and, to a lesser extent, strain and certain biological interactions (Williams, 1992; Silversides & Scott, 2001). If eggs from Muanda were measured sooner or stored under different conditions, the difference could be environmental or procedural rather than intrinsic to the hens. Tadesse et al. (2015) also showed that production system may be associated with differences in Haugh units, although the direction of the effect depends on strain and management conditions. Experimental evidence also shows that extended refrigerated storage can reduce albumen height and Haugh units (Jones & Musgrove, 2005). Consequently, the present territorial contrast cannot be attributed to geographical origin alone.

The strong correlation between albumen height and Haugh units was expected because height is the main positive term in the equation. The negative correlation between egg weight and Haugh units does not necessarily indicate that heavier eggs are of poor quality; it partly reflects the mathematical weight correction in the formula. Direct selection for both Haugh units and egg size should therefore account for their relationship and the measurement context.

4.3. Biological interpretation of the PCA

The first PCA component, explaining 53.25% of variance, can be interpreted as a size factor. Length and width contributed almost equally, with weight loading in the same direction. This structure is consistent with findings from Zambia, where Liswaniso et al. (2020) interpreted the first component as a size factor with high contributions from weight and dimensions. In

their study, three components explained 79.05% of variance; here, two components explained 79.60%, partly because fewer active variables were included.

The second component, dominated by Haugh units, separated internal quality from external size. PCA components are orthogonal by construction; this property does not establish biological independence between egg size and albumen quality (Jolliffe & Cadima, 2016). It theoretically allows the identification of hens combining eggs of acceptable size with high Haugh units. However, the observed component scores should first be confirmed in a longitudinal design that standardizes hen age, feeding, measurement timing, and storage conditions.

4.4. Scope of the typology

Clustering identified a majority group producing smaller eggs with slightly higher Haugh units and a minority group characterized by larger and heavier eggs. This contrast is biologically plausible from an animal-production perspective: the first profile may be valuable for internal quality, whereas the second may better satisfy commercial preferences for larger eggs. Nevertheless, the silhouette coefficient of 0.284 indicates overlap between groups. They should be considered provisional statistical profiles rather than definitively established biological types. Silhouette analysis evaluates cohesion and separation within the fitted sample; it does not substitute for external validation or stability assessment (Rousseeuw, 1987; Hennig, 2007).

The lack of a statistically significant association between group and territory ($\chi^2 = 0.609$; $p = 0.737$) indicates that the observed cluster membership was not demonstrably related to territory in this sample. Accordingly, the results do not support prioritizing one territory solely on the basis of this exploratory typology. Hens should instead be characterized individually over repeated laying cycles. No genetic inference can be drawn from this egg-based classification: phenotypic differentiation does not establish distinct breeds or genomic divergence.

4.5. Study limitations

The findings are limited to an exploratory sample of 38 hens from three territories. Although 190 eggs were measured, the principal territorial comparisons and typology used 38 hen-level means. Sample size reflected resources and collection feasibility rather than an a priori power calculation. Household and hen selection probabilities were not documented,

preventing a claim of probability-based representativeness. The veterinary-registry sampling frame, voluntary participation, and restriction to households with at least five actively laying hens further limit coverage and generalization, particularly to smaller flocks. Nonsignificant comparisons do not demonstrate equivalence or exclude smaller differences.

The study population is now characterized by age greater than 8 months, live body weight of 900–1,450 g, free-range scavenging based on household by-products and local feed resources, onset of laying at 6–8 months, and completion of at least two laying cycles. These criteria improve the description of eligible birds but do not establish uniform age, nutritional intake, or physiological stage. In particular, completion of two laying cycles does not specify the position within the cycle when eggs were collected. Detailed individual-level ages, weights, dietary intake, and within-cycle timing were not sufficiently documented in the manuscript to support covariate adjustment. Apparent health also does not constitute a standardized health assessment. Age, feeding, and health can influence egg traits, so their possible contributions remain unresolved (Roberts, 2004; Silversides & Scott, 2001).

Post-laying conditions remain a major source of uncertainty. The interval between laying and measurement, transport conditions, and storage temperature were not fully documented. Because albumen height and Haugh units change with storage, territorial differences may reflect handling as well as biological or husbandry variation (Williams, 1992; Jones & Musgrove, 2005). They cannot be assigned specifically to geographical origin or intrinsic hen quality.

Five consecutive eggs per hen ensured equal numerical contributions but did not remove within-hen dependence or establish repeatability across seasons and laying cycles. Hen-level means reduced pseudoreplication in the main analyses, whereas egg-level correlations and comparisons remain descriptive or exploratory (Hurlbert, 1984). Aggregation did not partition within-hen and between-hen variance; sampling one hen per household also prevented separation of hen and household effects. Some territory-by-cluster cells were small, warranting caution with the asymptotic chi-square test and future assessment using an exact or permutation procedure. The nonsignificant association does not prove independence of territory and cluster membership.

The typology was constructed and evaluated in the same sample without independent validation or reported resampling-based stability assessment. Its silhouette coefficient of 0.284 indicates limited separation in this dataset, consistent with provisional profiles along a phenotypic continuum (Rousseeuw, 1987). Cluster stability should be examined separately, because internal separation alone does not establish reproducible groups (Hennig, 2007). The measured traits do not establish overall laying productivity, shell quality, hatchability, or economic performance. Egg weight also enters the Haugh-unit equation, precluding interpretation of their association solely as a biological trade-off. Finally, phenotypic observations without pedigree, molecular data, or an appropriate genetic design cannot partition genetic and environmental variance or establish breeds, heritability, or expected selection response. Breeding and conservation applications require broader sampling, standardized measurements, longitudinal follow-up, and independent validation.

4.6. Research perspectives

The findings suggest several research priorities. First, sampling should be expanded to include more hens, households, and localities to confirm the stability of the observed typological profiles. Repeated measurements across several seasons and laying cycles should be incorporated to distinguish hen-specific variability from variation related to environmental conditions and management practices.

Future studies should also document hen age, parity, body weight, health status, diet, the interval between laying and measurement, and egg transport and storage conditions using standardized procedures. Additional traits such as yolk, albumen, and shell weights; shell thickness; yolk color; and yolk index would refine the typology. Finally, validating the groups in an independent sample and combining mixed-effects models with molecular markers, where possible, would help determine whether the observed profiles reflect simple phenotypic variation or genetic resources of potential value for breeding and conservation programs. Resampling can evaluate cluster stability, while an independent cohort is needed to assess whether the profiles recur outside the development sample (Hennig, 2007).

5. Conclusion

This study documented phenotypic variability among local hens from Mbanza-Ngungu, Muanda, and Songololo in egg morphometric traits and albumen

quality. Egg weight did not show a robust territorial difference at the hen level, whereas Haugh units differed among territories and were highest in Muanda. Because Haugh units are sensitive to measurement timing and storage conditions, this territorial contrast should be interpreted cautiously.

Using the hen as the primary statistical unit for the main comparisons reduced the risk of pseudoreplication arising from five eggs measured per hen. PCA and hierarchical clustering identified two exploratory profiles differentiated mainly by egg size and, secondarily, by Haugh units. Their limited separation and the absence of a detected association with territory indicate that these profiles should not be regarded as definitive biological or genetic categories.

The results provide a preliminary basis for individual phenotypic characterization of local hens in Kongo Central. Before the proposed typology can inform breeding or conservation decisions, it should be validated in larger and independent samples, with repeated measurements across seasons and laying cycles and standardized documentation of hen age, feeding, health status, egg handling, laying-to-measurement interval, and storage conditions. Where feasible, reproductive-performance and genomic data should be integrated to determine the biological and genetic relevance of the observed profiles.

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Conflict of Interest

The authors declare no conflicts of interest.

Ethical Considerations

Data were collected from chicken keepers in the study area after obtaining their informed consent. Participants were informed of the purpose and importance of the study before agreeing to participate.

Author Contributions

E.D.O. conceptualized the study, developed the study protocol, collected field data, and wrote the original manuscript draft.

A.T.T, C.G.T.M., G.K.B., and A.T.N. contributed to reviewing the study protocol, performing statistical analyses, and reviewing and editing the manuscript.

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