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# The effect of varying levels of corticated marama bean (*Tylosema esculentum*) meal on growth performance, apparent retention of feed components, and physiological and meat quality parameters in Jumbo quail

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A R T I C L E  I N F O

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A B S T R A C T

This study evaluated the effect of varying levels of corticated marama bean (*Tylosema esculentum*) meal (CMBM) on growth performance, apparent retention (AR) of feed components, and physiological and meat quality responses in Jumbo quail. Five isocaloric and isoproteic diets were formulated to contain 0 (T1), 51.25 (T2), 67.49 (T3), 83.74 (T4), and 99.98 g/kg (T5) of CMBM. A total of 385, one-week-old quail chicks (26.7 ± 2.16 g live-weight) were placed in cages (11 birds/cage) based on body weight. The diets were allocated to cages in a completely randomised design to give 7 replicate cages per diet. Feed intake (FI), body weight gain (BWG), gain-to-feed ratio (G:F), as well as AR of feed components were measured. The birds were slaughtered at day 35 for measurements of blood indices, organs weight, carcass and meat quality attributes. Feeding increasing levels of CMBM reduced FI ($P = 0.005$) linearly in week 2 but had no effect on FI in weeks 3–5. Similarly, linear reductions ($P < 0.0001$) were observed for average weekly BWG and G:F in weeks 2 and 3. A positive linear effect ($P = 0.033$) and a negative cubic effect ($P = 0.048$) for AR of neutral detergent fibre and organic matter were recorded as CMBM levels increased, respectively. However, varying responses ($P < 0.05$) were observed for AR of essential amino acids (methionine, lysine, threonine, etc.). Serum lipase, neutrophils, eosinophils, monocytes, and basophils linearly increased ($P < 0.05$) with dietary CMBM levels. Significant cubic or quartic effects ($P < 0.05$) were recorded for serum glucose, albumin, white blood cells, neutrophils, monocytes, basophils, and platelets. There were negative linear effects ($P < 0.05$) for final body weight, carcass yield, and breast meat drip loss. However, linear increases were recorded for gizzard, proventriculus, small intestine, and large intestine weights as dietary CMBM levels increased. In conclusion, the inclusion of CMBM up to 99.98 g/kg in Jumbo quail diets compromised growth performance, physiological responses, slaughter weights and carcass yield of Jumbo quail.

*Abbreviations:* AR, apparent retention; BWG, body weight gain; CCW, cold carcass weight; CMBM, corticated marama bean meal; FBW, final body weight; FI, feed intake; G:F, gain-to-feed ratio; HCW, hot carcass weight.

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## Introduction

Jumbo quail, belonging to *Coturnix* species, is an early-maturing bird known for its fast growth rates, high productivity and adaptability, short generation intervals, strong immunity, and low mortality rates, making it an ideal candidate to complement chicken production [1]. In South Africa, quail farming is an emerging agribusiness that has the potential to contribute high-quality animal protein to the food value chain [2]. Quail meat is popular amongst health-conscious consumers due to its high protein-to-fat ratio, low cholesterol levels, leanness, and low intramuscular fat [3]. The eggs are also commended for their high concentrations of fat soluble and B-complex vitamins, essential amino acids, as well as low levels of triglycerides, cholesterol, and saturated fatty acids [4]. However, sustainable quail farming is currently limited by the over-reliance on soybean meal as a major protein source in commercial animal diets [5].

Globally, the high demand for soybean for human and animal consumption as well as its numerous industrial applications have led to a rise in its prices [6]. Further, the production of soybean in South Africa does not meet the domestic demand due to various challenges including limited arable land, droughts, and pests, among others [7]. Thus, the reliance on foreign markets to meet the local demand for soybean poses significant economic sustainability challenges for quail farming in South Africa [8]. This calls for the scientific exploration of naturally growing legume crops that do not require human intervention, arable land, gallons of water, fertilisers, pesticides, and heavy machinery, as feed ingredients in quail diets [9].

Marama bean (*Tylosema esculentum*, family: *Fabaceae*) plant is a seasonal drought-tolerant tuberous legume [10] that has little-to-no direct food value to the South African general citizenry. This legume is native to Southern Africa where it is adaptable to the

**Table 1**
Ingredients and chemical composition of experimental diets containing corticated marama bean (*Tylosema esculentum*) meal (CMBM).

| | | [a]Diets | | | | |
|---|---|---|---|---|---|---|
| Ingredients (g/kg as fed) | CMBM | T1 | T2 | T3 | T4 | T5 |
| CMBM | | 0 | 51.25 | 67.49 | 83.74 | 99.98 |
| Yellow maize | | 552 | 558 | 564 | 574 | 585 |
| Full fat soybean meal | | 120 | 120 | 120 | 84.4 | 28.8 |
| Soya oilcake (46 %) | | 267 | 218 | 204 | 216 | 245 |
| Limestone powder | | 12.4 | 12.4 | 12.3 | 12.1 | 12.0 |
| Monocalcium phosphate | | 8.98 | 9.17 | 9.36 | 9.64 | 9.96 |
| Fine salt | | 2.71 | 2.65 | 2.50 | 2.53 | 2.48 |
| Sodium bicarbonate | | 1.27 | 1.37 | 1.46 | 1.55 | 1.63 |
| DL-Methionine | | 4.14 | 4.22 | 4.30 | 4.36 | 4.42 |
| Lysine-HCl | | 2.85 | 3.11 | 3.36 | 3.60 | 3.82 |
| L-Threonine | | 1.12 | 1.18 | 1.23 | 1.27 | 1.30 |
| L-Tryptophan | | 0.09 | 0.08 | 0.06 | 0.06 | 0.05 |
| Soybean oil | | 21.0 | 12.1 | 3.20 | 0 | 0 |
| [b]Lignobond | | 2.0 | 2.0 | 2.0 | 2.0 | 2.0 |
| [c]Premix | | 3.0 | 3.0 | 3.0 | 3.0 | 3.0 |
| Axtra phytase (10,000 FTU/g) | | 0.10 | 0.10 | 0.10 | 0.10 | 0.10 |
| Antioxidant | | 0.25 | 0.25 | 0.25 | 0.25 | 0.25 |
| Salinomycin (12 %) | | 0.50 | 0.50 | 0.50 | 0.50 | 0.50 |
| Zinc bacitracin | | 0.50 | 0.50 | 0.50 | 0.50 | 0.50 |
| Composition (g/kg DM, unless stated otherwise) | | | | | | |
| Calculated ME[d] (Kcal/kg) | 2750 | 2774 | 2772 | 2772 | 2773 | 2771 |
| Dry matter (g/kg) | 929 | 906 | 906 | 912 | 910 | 913 |
| Organic matter | 903 | 860 | 864 | 869 | 866 | 865 |
| Crude protein | 264 | 235 | 235 | 236 | 235 | 235 |
| Crude fibre | 486 | 62.6 | 99.9 | 102 | 93.4 | 123 |
| Ether extract | 247 | 35.9 | 48.2 | 52.2 | 43.8 | 43.0 |
| Neutral detergent fibre | 501 | 111 | 135 | 144 | 152 | 158 |
| Acid detergent fibre | 476 | 47.7 | 54.9 | 58.9 | 58.4 | 56.3 |
| Acid detergent lignin | 207 | 2.26 | 2.47 | 4.73 | 4.76 | 9.35 |
| Calcium | 0 | 56.6 | 36.0 | 38.4 | 39.0 | 33.0 |
| Phosphorus | 0.51 | 2.69 | 2.32 | 2.37 | 2.48 | 2.45 |
| Threonine (g /100 g DM) | 1.44 | 1.81 | 1.19 | 1.19 | 1.86 | 1.29 |
| Lysine (g /100 g DM) | 2.11 | 2.42 | 2.00 | 1.48 | 1.47 | 2.38 |
| Methionine (g /100 g DM) | 0.39 | 0.64 | 0.91 | 0.65 | 0.97 | 0.60 |
| Leucine (g /100 g DM) | 2.62 | 3.39 | 2.60 | 2.52 | 3.25 | 2.60 |

[a] Diets: T1 = *a* conventional grower diet without CMBM; T2 = *a* conventional grower diet containing 51.25 g/kg CMBM; T3 = *a* conventional grower diet containing 67.49 g/kg CMBM; T4 = *a* conventional grower diet containing 83.74 g/kg CMBM; T5 = *a* conventional grower diet containing 99.98 g/kg CMBM.

[b] Lignobond = high performance lignin-based pellet binders.

[c] Premix: vitamin A (11,000 IU), vitamin B1 (2.5 mg), vitamin B2 (4.5 mg), vitamin B6 (5.1 mg), vitamin D3 (2500 IU), vitamin E (25 IU), vitamin K3 (2.0 mg) biotin (0.12 g), folic acid (0.7 mg), niacin (30 mg), pantothenic acid (10 mg).

[d] ME = metabolizable energy.

All the ingredients were received from Nutroteq and SimpleGrow Agric Services (Centurion, South Africa).

tropical and sub-tropical environmental conditions characterised by low rainfall and poor-quality soils [11]. The marama bean has high levels of crude protein (36 – 37%) and oil (24 – 48%) that are comparable to that of soybean [11]. It is also abundant in calcium, zinc, phosphorus, potassium, iron, magnesium, and vitamins A and B-complex [10]. However, it also contains high concentrations of trypsin inhibitors and condensed tannins [12], which could limit its utilisation in Jumbo quail diets.

In addition, the seed is naturally covered with a hard coat that is laborious and expensive to decorticate, which could be the reason it is neglected and underutilised in animal feeds. Unfortunately, the use of corticated marama bean meal (CMBM) in Jumbo quail diets could increase dietary fibre levels resulting in poor utilisation, however, to the authors' knowledge, no studies have tested feeding value of CMBM. Thus, this study investigated the effect of including varying levels of CMBM in a Jumbo quail diet on growth performance, apparent retention (AR) of feed components, and physiological and meat quality parameters.

## Materials and methods

### *Processing and analyses of corticated marama bean*

Corticated full-fat marama beans were procured from street vendors in Malwelwe village (23.94282° S; 25.1999° E; Kweneng, Botswana). The beans were crushed using a Trapp forage shredder (model number: TFR 400) in Benoni (Gauteng, South Africa) to produce CMBM. The meal was further homogenised (2 mm) using a Polymix grinder (model no: PX-MFC 90D, Switzerland) for chemical analyses and diet formulation.

The CMBM samples were analysed for dry matter (DM; method no. 930.15), organic matter (OM; method no. 942.05), ether extract (EE; method no. 920.39), and crude protein (CP; method no. 984.13) following AOAC guidelines [13]. Amino acids (leucine, lysine, methionine, and threonine) were analysed at Stellenbosch University (Western Cape, South Africa) using the Waters Acquity UPLC fitted with a photodiode array detector as described by Chikwanha et al. [14]. The automated ANKOM$^{DELTA}$ Fibre Analyser (ANKOM Technology, New York, USA) was used to determine crude fibre content (AOCS method Ba 6a-05) after digesting with sulphuric acid and sodium hydroxide for 80 min. Neutral detergent fibre (NDF) and acid detergent fibre (ADF) were assayed after refluxing pre-weighed samples with neutral and acid detergent solutions for 60 min and 75 min, respectively, using the same automated ANKOM$^{DELTA}$ Fibre Analyser. The analysis of the NDF involved the use of α-amylase and was expressed inclusive of residual ash [15]. The ADF residue bags were submerged using 72% $H_2SO_4$ for 3 h and oven dried at 105°C overnight to determine acid detergent lignin (ADL). Metabolizable energy (ME) was calculated as: ME (kcal/kg) = 2778 – 66 (Crude fibre × 88% DM).

### *Diet formulation and analysis*

Five experimental diets were formulated (Table 1) to meet the nutritional requirements of *Coturnix* broiler quail [16] by adding 0 (T1), 51.25 (T2), 67.49 (T3), 83.74 (T4), and 99.98 g/kg (T5) of CMBM in a conventional grower diet. The diets were in a mash form (2 mm) and were isonitrogenous and isocaloric. The chemical constituents and ME values of the diets were determined as described for the CMBM above.

### *Feeding trial and performance measurements*

The care, rearing and slaughtering of the birds was approved (NWU-01884-19-S5) by the North-West University's Research Ethics Committee for Animal Studies (Mafikeng, South Africa). The experiment was conducted between July and August 2022 at the North-West University research farm (25°34′58″ S, 25°64′32″ E; South Africa). In the same house, 385 one-week-old unsexed Jumbo quail chicks were weighed and randomly allocated into 35 replicate cages (11 birds/cage). The five experimental diets were allocated to cages using completely randomized design to give 7 replicate cages per diet. The cages (100 cm long × 60 cm wide × 30 cm high) were built using wire-mesh and the floors were completely covered using polythene sheets. The birds were offered a Phenix stress pack (consisting of multi-vitamins and electrolytes) in fresh water for the first 3 days. Thereafter, fresh clean drinking water and feed were offered *ad libitum* until termination. The feed was provided using Poltek poultry oval-hole feeder (4/100, NWK, Mafikeng, South Africa) and the water was provided using Poltek poultry water fountain (3 L 3/10, NWK, Mafikeng, South Africa). Infrared lights were used to maintain house temperature at 35 °C for the first two weeks and thereafter removed for reliance on ambient temperature. The humidity of the house was maintained at an average of 65% and the house was ventilated by opening windows in the morning (6 am) and closing them in the evening (6 pm). Average feed intake (FI) per pen was measured daily as the difference between feed offered and refusals. All the birds per cage were weighed on day 7 (26.7 ± 2.16 g initial BW) and thereafter weighed weekly until day 35 to determine average weekly body weight gain (BWG). The FI and BWG data were computed to determine gain-to-feed ratio (G:F). The weight of dead birds was recorded on occurrence to determine mortality rates and the data was used to calculate G:F.

### *Apparent retention of feed components*

At day 35, final body weights were recorded and then two birds were randomly selected and left in the cages ($n = 70$) to allow for excreta collection. The birds were offered the same experimental diets and adapted for three days. The birds were then fasted for overnight before measurements (feed intake and excreta) were taken for five days. The excreta samples were weighed and pooled before being oven-dried (55 °C) to constant weight. The samples were thereafter milled (1 mm) and analysed using the methods described for the CMBM in Section 2.1. Apparent retention (AR) of DM, OM, CP, NDF and essential amino acids (arginine, histidine, lysine, isoleucine, leucine, methionine, phenylalanine, threonine, and valine) were determined using the formula:

$$Apparent\ retention = \frac{(Component\ intake - Component\ excreted)}{Component\ intake} \times 100$$

*Slaughter protocol, and blood collection and analysis*

All birds (apart from those subjected to the retention trial) were stunned and exsanguinated by slicing the jugular vein with a sharp knife at a local poultry abattoir. The blood of 2 randomly selected birds per experimental unit was allowed to drip into sterilised containers, from which blood samples (4 mL) were immediately collected using 5 mL syringes and transferred into sterilized serum biochemical and haematological (with ethylenediaminetetraacetic acid) tubes using 23 g needles. The clotted blood in serum tubes was centrifuged at 2659.95 RCF (Cryste Varispin 4 Multi-purpose centrifuge, Cryste CO LTD, Korea) to generate serum [17], which was analysed for albumin-to-globulin ratio (ALB/GLOB), albumin, symmetric dimethylarginine (SDMA), gamma-glutamyl transferase (GGT), alkaline phosphatase (ALKP), amylase, cholesterol, calcium, globulin, glucose, lipase, phosphorus, and total protein using an automated Catalyst One Chemistry Analyser (IDEXX Laboratories (Pty) Ltd., Gauteng, South Africa). Haematological parameters viz., haemoglobin, white blood cells (WBC), haematocrit, mean corpuscular haemoglobin (MCH), mean corpuscular volume (MCV), lymphocytes, red cell distribution width (RDW), platelet, basophils, neutrophils, monocytes, and eosinophils were determined using an automated LaserCyte Haematology Analyser (IDEXX Laboratories (Pty) Ltd., Gauteng, South Africa).

*Carcass characteristics and internal organs*

The carcasses (60 – 63 per treatment) were plucked and eviscerated upon slaughter and immediately weighed to determine hot carcass weight (HCW). The carcasses were then stored at 2 °C for 24 h and thereafter re-weighed to determine cold carcass weights (CCW). Carcass yield (%) was determined as a proportion of CCW to final body weight (FBW). The carcasses were cut into retail parts to measure breast, drumstick, thigh, and wing weights using a digital scale (Explorer EX224, 0.01 g readability, OHAUS Corp., Parsippany, NJ, USA). The weights of the gizzards, spleen, liver, proventriculus, and small and large intestines were also measured.

*Breast meat quality measurements*

Breast meat pH was measured 24 h postmortem around the left, middle and right sides of the breast from all the carcasses using a pH metre (HI98163 pH metre, Hanna Instruments (Pty) Ltd, JHB, South Africa). The metre was calibrated using conventional standard solutions (pH 4, 7 and 10) for every experimental unit. The lightness ($L^*$), yellowness ($b^*$) and redness ($a^*$) were also recorded using a colorimeter (Konica Minolta Chroma metre CR-400, Narich (Pty) Ltd, Japan) set to read at an illuminant D65-day light, 20 mm diameter measurement area, and 10° standard observer. Meat colour was measured in three positions of the upper surface of each breast fillet. The triplicate values for meat pH and colour were averaged before being subjected to statistical analysis. Drip loss was assayed after hanging pre-weighed samples in air-tight plastic bottles which were stored in a cold room at 2 °C for 72 h [18,19]. Water holding capacity was evaluated by placing pre-weighed breast meat samples in-between filter papers and pressed with an external

**Table 2**
Impact of different levels of corticated marama bean (*Tylosema esculentum*) meal on overall mortality (%) and growth performance in Jumbo quail.

| | [a]Diets | | | | | | Significance | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| [b]Metrices | T1 | T2 | T3 | T4 | T5 | [c]SEM | Linear | Quadratic | Cubic | Quartic |
| Overall mortality (%) | 2.60 | 12.88 | 11.7 | 15.7 | 8.87 | 3.415 | 0.010 | 0.444 | 0.493 | 0.644 |
| Average weekly feed intake (g/bird) | | | | | | | | | | |
| FI_Wk 2 | 80.9 | 73.2 | 75.5 | 69.1 | 70.9 | 2.685 | 0.005 | 0.401 | 0.825 | 0.124 |
| FI_Wk 3 | 119 | 118 | 119 | 114 | 116 | 3.659 | 0.333 | 0.991 | 0.726 | 0.513 |
| FI_Wk 4 | 147 | 151 | 156 | 150 | 155 | 3.518 | 0.161 | 0.858 | 0.426 | 0.210 |
| FI_Wk 5 | 199 | 206 | 213 | 200 | 205 | 4.34 | 0.555 | 0.322 | 0.249 | 0.114 |
| Average weekly body weight gain (g/bird) | | | | | | | | | | |
| WG_Wk 2 | 41.4 | 35.2 | 32.9 | 25.8 | 24.1 | 1.448 | 0.0001 | 0.734 | 0.725 | 0.126 |
| WG_Wk 3 | 54.1 | 53.2 | 46.9 | 41.9 | 37.9 | 1.584 | 0.0001 | 0.112 | 0.214 | 0.599 |
| WG_Wk 4 | 72.8 | 69.3 | 65.7 | 63.5 | 60.3 | 9.735 | 0.306 | 0.986 | 0.972 | 0.962 |
| WG_Wk 5 | 27.9 | 32.7 | 34.4 | 46.7 | 39.8 | 7.040 | 0.104 | 0.820 | 0.478 | 0.468 |
| Average weekly gain-to-feed ratio | | | | | | | | | | |
| G:F_Wk 2 | 0.513 | 0.487 | 0.435 | 0.371 | 0.340 | 0.013 | 0.0001 | 0.118 | 0.250 | 0.832 |
| G:F_Wk 3 | 0.453 | 0.449 | 0.394 | 0.368 | 0.329 | 0.064 | 0.0001 | 0.112 | 0.357 | 0.258 |
| G:F_Wk 4 | 0.494 | 0.456 | 0.423 | 0.427 | 0.388 | 0.034 | 0.220 | 0.992 | 0.812 | 0.845 |
| G:F_Wk 5 | 0.139 | 0.157 | 0.160 | 0.232 | 0.195 | 0.014 | 0.101 | 0.954 | 0.383 | 0.363 |

[a] Diets: T1 = *a* conventional grower diet without CMBM; T2 = *a* conventional grower diet containing 51.25 g/kg CMBM; T3 = *a* conventional grower diet containing 67.49 g/kg CMBM; T4 = *a* conventional grower diet containing 83.74 g/kg CMBM; T5 = *a* conventional grower diet containing 99.98 g/kg CMBM.

[b] Metrices: FI = feed intake; BWG = body weight gain; G:*F* = gain-to-feed ratio.

[c] SEM = standard error of the mean.

pressure of 60 kg for 5 min [20].

*Statistical analysis*

The data for growth metrices, AR of feed components, blood parameters, carcass traits, visceral organs, and meat quality were examined for normality using PROC Univariate in SAS version 9.4 [21]. Polynomial contrasts were used to evaluate the data for linear, quadratic, cubic and quartic responses in SAS. The optimum inclusion level of CMBM was determined using response surface regression analysis (PROC RSREG) using the quadratic equation:

$$y = c + bx + ax^2$$

Where $y$ is the response variable, $a$ and $b$ are the coefficients of the quadratic equation, $c$ is the intercept and $x$ is CMBM level (g/kg). Optimal responses were calculated as $x = -b/2a$. The level of significance was set at $P < 0.05$ for all the tested parameters.

## Results

Table 1 indicates that, although the diets were formulated to have the same levels of ME and CP, the increase in dietary CMBM levels tended to increase the fibre (crude fibre, NDF, ADF and ADL) levels of the diets. Feeding diets with varying levels of CMBM linearly increased ($P = 0.01$) overall mortality rates (%) as shown in Table 2. In week 2, feeding diets with varying levels of CMBM resulted in a negative linear effect ($P = 0.005$) for FI. Similarly, negative linear responses ($P < 0.0001$) were observed for average weekly BWG and G:F in weeks 2 and 3 in response to dietary CMBM levels. No quadratic, cubic or quartic responses ($P > 0.05$) were recorded for overall mortality and weekly growth performance parameters in response to incremental levels of dietary CMBM.

No linear, quadratic, cubic or quartic responses ($P > 0.05$) were observed for the AR of DM and CP (Table 3). However, a positive linear effect ($P = 0.033$) was recorded for AR of NDF, while a negative cubic response ($P = 0.048$) was noted for AR of OM as dietary CMBM levels increased. The AR of arginine, histidine, isoleucine, leucine, and valine decreased linearly ($P < 0.05$) to CMBM levels. Additionally, quadratic effects ($P < 0.05$) were observed for AR of arginine, isoleucine, leucine, lysine, and valine with dietary CMBM levels. Moreover, feeding diets with varying levels of CMBM caused cubic responses ($P < 0.05$) for the AR of arginine, histidine, isoleucine, leucine, methionine, phenylalanine, threonine, and alanine. Lastly, quartic effects ($P < 0.05$) were recorded for AR of arginine, isoleucine, methionine, and phenylalanine as CMBM levels increased.

No linear or quadratic effects were recorded for serum biochemical parameters, apart from serum lipase ($P = 0.001$) which linearly increased with dietary CMBM levels (Table 4). Also, feeding diets with varying levels of dietary CMBM caused cubic and quartic responses ($P > 0.05$) for serum glucose and albumin levels, respectively.

Inclusion of varying levels of CMBM linearly increased the concentrations of neutrophils ($P = 0.017$), eosinophils ($P = 0.028$), monocytes ($P = 0.043$), and basophils ($P = 0.025$) in Jumbo quail (Table 5). Significant cubic effects were recorded for white blood cells (WBC), neutrophils, monocytes, basophils, and platelets in response to different levels of CMBM.

There were negative linear effects for FBW ($P = 0.012$), carcass yield ($P = 0.035$), and breast meat drip loss ($P = 0.01$) as dietary CMBM levels increased (Table 6). However, positive linear increases were recorded for gizzard ($P = 0.0001$), proventriculus ($P = 0.049$), small intestine ($P = 0.0001$) and large intestine weights ($P = 0.037$). There were both linear and quadratic responses for HCW [$y = 141\ (\pm 3.827) + 0.398\ (\pm 0.433)\ x - 0.036\ (\pm\ 0.011)\ x^2$; $R^2 = 0.722$, $P = 0.004$] and CCW [$y = 140\ (\pm 3.792) + 0.449\ (\pm 0.429)\ x -$

**Table 3**
Apparent retention of proximate components (%) and essential amino acids (% DM) in Jumbo quail fed with different levels of corticated marama (*Tylosema esculentum*) bean meal.

| | [a]Diets | | | | | | Significance | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| Components | T1 | T2 | T3 | T4 | T5 | [c]SEM | Linear | Quadratic | Cubic | Quartic |
| Dry matter | 60.9 | 61.3 | 64.9 | 62.1 | 63.1 | 0.310 | 0.605 | 0.669 | 0.963 | 0.463 |
| Organic matter | 5.96 | 5.28 | 6.30 | 5.76 | 4.90 | 0.321 | 0.104 | 0.102 | 0.048 | 0.087 |
| [b]NDF | 19.7 | 27.1 | 30.8 | 34.2 | 34.2 | 5.020 | 0.033 | 0.408 | 0.978 | 0.865 |
| Crude protein | 31.8 | 27.6 | 36.7 | 30.2 | 31.6 | 4.852 | 0.906 | 0.830 | 0.761 | 0.264 |
| Arginine | 91.8 | 82.7 | 81.5 | 88.7 | 821 | 1.276 | 0.002 | 0.009 | 0.0001 | 0.042 |
| Histidine | 94.8 | 87.0 | 87.7 | 91.7 | 83.3 | 0.988 | 0.0001 | 0.510 | 0.0001 | 0.212 |
| Isoleucine | 88.2 | 78.5 | 75.4 | 84.0 | 78.4 | 1.736 | 0.015 | 0.005 | 0.001 | 0.043 |
| Leucine | 89.9 | 82.4 | 81.8 | 86.0 | 81.7 | 1.387 | 0.007 | 0.035 | 0.002 | 0.341 |
| Lysine | 90.9 | 84.7 | 79.2 | 80.5 | 87.6 | 1.657 | 0.052 | 0.0001 | 0.343 | 0.622 |
| Methionine | 86.2 | 86.2 | 82.0 | 90.3 | 83.1 | 1.393 | 0.657 | 0.734 | 0.015 | 0.001 |
| Phenylalanine | 88.9 | 85.0 | 85.1 | 90.7 | 845. | 1.170 | 0.382 | 0.826 | 0.0001 | 0.065 |
| Threonine | 86.2 | 71.3 | 75.3 | 84.2 | 76.6 | 1.927 | 0.298 | 0.012 | 0.0001 | 0.660 |
| Valine | 87.7 | 76.5 | 74.6 | 83.1 | 76.0 | 1.832 | 0.007 | 0.011 | 0.0001 | 0.083 |

[a] Diets: T1 = *a* conventional grower diet without CMBM; T2 = *a* conventional grower diet containing 51.25 g/kg CMBM; T3 = *a* conventional grower diet containing 67.49 g/kg CMBM; T4 = *a* conventional grower diet containing 83.74 g/kg CMBM; T5 = *a* conventional grower diet containing 99.98 g/kg CMBM.
[b] NDF = neutral detergent fibre.
[c] SEM = standard error of the mean.

**Table 4**
Impact of different levels of corticated marama bean (*Tylosema esculentum*) meal on serum biochemical parameters in five-week-old Jumbo quail.

| [2]Parameter | [1]Diets | | | | | [3]SEM | Significance | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| | T1 | T2 | T3 | T4 | T5 | | Linear | Quadratic | Cubic | Quartic |
| Glucose (mmol/L) | 15.1 | 15.0 | 14.7 | 13.5 | 14.7 | 0.364 | 0.081 | 0.335 | 0.028 | 0.187 |
| SDMA (µg/dL) | 30.9 | 32.2 | 30.3 | 30.7 | 34.8 | 2.420 | 0.701 | 0.696 | 0.368 | 0.845 |
| Phosphorus (mmol/L) | 2.78 | 3.00 | 2.74 | 3.24 | 2.60 | 0.220 | 0.836 | 0.632 | 0.361 | 0.097 |
| Calcium (mmol/L) | 2.88 | 3.12 | 2.72 | 2.97 | 2.59 | 0.195 | 0.256 | 0.701 | 0.984 | 0.123 |
| Total protein (g/L) | 36.6 | 39.2 | 36.5 | 41.9 | 35.0 | 2.435 | 0.780 | 0.630 | 0.366 | 0.106 |
| Albumin (g/L) | 11.4 | 12.6 | 11.5 | 12.1 | 11.4 | 0.362 | 0.543 | 0.397 | 0.459 | 0.028 |
| Globulin (g/L) | 25. | 26.7 | 25.3 | 29.7 | 23.7 | 2.178 | 0.810 | 0.528 | 0.294 | 0.177 |
| ALB/GLOB | 0.471 | 0.467 | 0.479 | 0.457 | 0.486 | 0.018 | 0.533 | 0.810 | 0.159 | 0.316 |
| ALKP (U/L) | 297 | 393 | 349 | 413 | 357 | 48.15 | 0.334 | 0.583 | 0.892 | 0.251 |
| GGT (U/L) | 2.64 | 4.43 | 4.71 | 3.57 | 2.86 | 1.312 | 0.847 | 0.134 | 0.645 | 0.872 |
| Bilirubin (µmol/L) | 5.00 | 8.00 | 5.71 | 6.64 | 4.57 | 1.494 | 0.843 | 0.461 | 0.632 | 0.249 |
| Cholesterol (mmol/L) | 4.28 | 4.80 | 4.56 | 4.79 | 4.44 | 0.192 | 0.236 | 0.314 | 0.770 | 0.167 |
| Amylase (U/L) | 211 | 238 | 208 | 274 | 198 | 32.35 | 0.926 | 0.307 | 0.424 | 0.157 |
| Lipase (U/L) | 325 | 409 | 561 | 648 | 675 | 65.45 | 0.001 | 0.494 | 0.541 | 0.805 |

[a] Diets: T1 = *a* conventional grower diet without CMBM; T2 = *a* conventional grower diet containing 51.25 g/kg CMBM; T3 = *a* conventional grower diet containing 67.49 g/kg CMBM; T4 = *a* conventional grower diet containing 83.74 g/kg CMBM; T5 = *a* conventional grower diet containing 99.98 g/kg CMBM.
[b] Parameters: ALKP = alkaline phosphatase; SDMA = symmetric dimethylarginine; ALB/GLOB = albumin-to-globulin ratio; GGT = gamma-glutamyl transferase.
[c] SEM: = standard error of the mean.

**Table 5**
Impact of different levels of corticated marama bean (*Tylosema esculentum*) meal on haematological parameters in five-week-old Jumbo quail.

| [b]Parameters | [a]Diet | | | | | [c]SEM | Significance | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| | T1 | T2 | T3 | T4 | T5 | | Linear | Quadratic | Cubic | Quartic |
| RBC ($\times 10^{12}$/L) | 2.87 | 2.45 | 2.66 | 2.22 | 2.96 | 0.252 | 0.922 | 0.214 | 0.405 | 0.204 |
| Haemoglobin (g/dL) | 11.3 | 12.1 | 11.3 | 11.5 | 11.7 | 0.655 | 0.465 | 0.775 | 0.355 | 0.435 |
| MCH (pg) | 43.1 | 52.4 | 43.9 | 46.3 | 40.5 | 3.634 | 0.462 | 0.151 | 0.306 | 0.665 |
| MCV (fL) | 81.2 | 76.0 | 84.2 | 83.3 | 84.4 | 2.782 | 0.218 | 0.450 | 0.138 | 0.274 |
| Haematocrit (%) | 24.2 | 23.1 | 23.8 | 21.5 | 24.8 | 1.744 | 0.393 | 0.381 | 0.884 | 0.251 |
| RDW (%) | 20.1 | 19.4 | 20.4 | 20.0 | 20.4 | 0.479 | 0.640 | 0.189 | 0.187 | 0.297 |
| Platelet (%) | 19.6 | 18.0 | 22.0 | 18.8 | 18.8 | 1.951 | 0.212 | 0.184 | 0.236 | 0.320 |
| Reticulocytes ($\times 10^9$/L) | 18.1 | 9.62 | 28.7 | 13.6 | 15.8 | 7.002 | 0.931 | 0.559 | 0.257 | 0.895 |
| WBC ($\times 10^9$/L) | 16.5 | 23.8 | 13.9 | 16.0 | 23.8 | 3.257 | 0.158 | 0.111 | 0.207 | 0.113 |
| Neutrophils ($\times 10^9$/L) | 5.6 | 8.4 | 6.1 | 7.9 | 9.8 | 1.000 | 0.017 | 0.374 | 0.158 | 0.136 |
| Lymphocytes ($\times 10^9$/L) | 7.56 | 7.32 | 4.18 | 5.12 | 8.89 | 1.335 | 0.850 | 0.273 | 0.109 | 0.159 |
| Monocytes ($\times 10^9$/L) | 2.72 | 2.76 | 2.95 | 2.37 | 4.14 | 0.309 | 0.043 | 0.229 | 0.595 | 0.182 |
| Eosinophils ($\times 10^9$/L) | 0.508 | 0.572 | 0.471 | 0.576 | 0.825 | 0.090 | 0.028 | 0.527 | 0.102 | 0.205 |
| Basophils ($\times 10^9$/L) | 0.099 | 0.116 | 0.096 | 0.103 | 0.155 | 0.012 | 0.025 | 0.165 | 0.391 | 0.228 |

[a] Diets: T1 = *a* conventional grower diet without CMBM; T2 = *a* conventional grower diet containing 51.25 g/kg CMBM; T3 = *a* conventional grower diet containing 67.49 g/kg CMBM; T4 = *a* conventional grower diet containing 83.74 g/kg CMBM; T5 = *a* conventional grower diet containing 99.98 g/kg CMBM.
[b] Parameters = MCH = mean corpuscular haemoglobin; MCV = mean corpuscular volume; RBC = red blood cells; RDW = red blood cell distribution width; WBC = white blood cells.
[c] SEM = standard error of the mean.

0.037 (±0.011) $x^2$; $R^2 = 0.723$, $P = 0.003$], which were optimized at 5.50 and 6.07 g CMBM /kg diet, respectively.

## Discussion

### *Performance and apparent retention of feed components*

The nutritive value of CMBM shows that its utilisation as a novel protein source could be limited by higher levels of non-starch polysaccharides (NSP), trypsin inhibitors and condensed tannins [22]. In this study, the inclusion of CMBM up to 99.98 g/kg reduced feed intake in two-week-old quail, which is attributable to the extra dietary fibre levels in the diets containing CMBM. Indeed, poultry birds have limited ability to utilise highly fibrous diets due to a lack of digestive fibrolytic enzymes (non-starch polysaccharidases). As such, dietary fibre forms a physical barrier against endogenous digestive enzymes, resulting in reduced feed digestion [23]. Thus, the significant decline in feed consumption could have been influenced by poor gut development in young quail compared to the 3 - 5-week-old birds [24]. The poor utilisation of the diets resulted in reduced BWG and G:F in weeks 2 and 3. This

**Table 6**
Impact of different levels of corticated marama bean (*Tylosema esculentum*) meal on carcass characteristics, breast meat quality, and internal organs in five-week-old Jumbo quail.

| | [1]Diets | | | | | | Significance | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| [2]Parameters | T1 | T2 | T3 | T4 | T5 | [3]SEM | Linear | Quadratic | Cubic | Quartic |
| Carcass yield (%) | 63.6 | 65.5 | 62.4 | 61.5 | 57.2 | 1.919 | 0.035 | 0.062 | 0.790 | 0.452 |
| FBW (g) | 223 | 217 | 204 | 204 | 189 | 7.136 | 0.012 | 0.530 | 0.728 | 0.440 |
| HCW (g) | 142 | 142 | 127 | 123 | 107 | 3.175 | 0.0001 | 0.004 | 0.834 | 0.075 |
| CCW (g) | 141 | 140 | 126 | 123 | 106 | 3.150 | 0.0001 | 0.003 | 0.958 | 0.081 |
| Drumstick (% HCW) | 4.37 | 4.41 | 4.48 | 4.35 | 4.59 | 0.145 | 0.953 | 0.506 | 0.457 | 0.526 |
| Thigh (% HCW) | 7.52 | 7.37 | 7.30 | 7.31 | 7.28 | 0.311 | 0.543 | 0.205 | 0.901 | 0.950 |
| Wing (% HCW) | 4.70 | 4.79 | 4.69 | 4.66 | 5.24 | 0.145 | 0.099 | 0.087 | 0.097 | 0.842 |
| Breast (% HCW) | 18.4 | 18.7 | 17.7 | 17.2 | 19.2 | 0.944 | 0.805 | 0.662 | 0.198 | 0.977 |
| pH | 5.98 | 6.06 | 6.09 | 6.07 | 6.07 | 0.044 | 0.187 | 0.316 | 0.741 | 0.645 |
| *L** (lightness) | 34.3 | 34.9 | 34.6 | 35.6 | 35.2 | 0.447 | 0.595 | 0.660 | 0.753 | 0.165 |
| *a**(redness) | 9.3 | 22.9 | 9.17 | 7.85 | 8.26 | 6.176 | 0.546 | 0.305 | 0.147 | 0.336 |
| *b** (yellowness) | 2.44 | 2.67 | 2.75 | 2.84 | 2.94 | 0.138 | 0.113 | 0.539 | 0.701 | 0.881 |
| WHC (%) | 88.1 | 86.7 | 88.2 | 86.5 | 86.1 | 1.657 | 0.711 | 0.894 | 0.765 | 0.446 |
| Drip loss (%) | 22.2 | 23.1 | 18.8 | 19.2 | 19.5 | 0.648 | 0.010 | 0.239 | 0.017 | 0.012 |
| Proventriculus (% HCW) | 0.619 | 0.723 | 0.641 | 0.687 | 0.735 | 0.041 | 0.049 | 0.577 | 0.159 | 0.213 |
| Gizzard (% HCW) | 2.44 | 2.89 | 2.96 | 3.09 | 3.41 | 0.085 | 0.0001 | 0.830 | 0.044 | 0.626 |
| Spleen (% HCW) | 0.096 | 0.154 | 0.179 | 0.169 | 0.119 | 0.045 | 0.135 | 0.413 | 0.969 | 0.990 |
| Liver (% HCW) | 0.029 | 0.029 | 0.030 | 0.030 | 0.032 | 0.002 | 0.196 | 0.534 | 0.675 | 0.742 |
| Small intestine (% HCW) | 4.03 | 4.52 | 5.42 | 4.84 | 5.18 | 0.351 | 0.0001 | 0.647 | 0.649 | 0.151 |
| Large intestine (% HCW) | 0.932 | 0.964 | 1.056 | 1.043 | 1.162 | 0.070 | 0.037 | 0.933 | 0.753 | 0.498 |

[a] Diets: T1 = *a* conventional grower diet without CMBM; T2 = *a* conventional grower diet containing 51.25 g/kg CMBM; T3 = *a* conventional grower diet containing 67.49 g/kg CMBM; T4 = *a* conventional grower diet containing 83.74 g/kg CMBM; T5 = *a* conventional grower diet containing 99.98 g/kg CMBM.
[b] Parameters: FBW = final body weight; HCW = hot carcass weight; CCW = cold carcass weight; WHC = water holding capacity.
[c] SEM = standard error of the mean.

further indicates that the extra dietary fibre in the CMBM treatments compromised the performance of the birds. Moreover, high lignin level in the marama bean coat could have been responsible for diluting digestible cell wall components since it is inherently indigestible, resulting in poor feed efficiency. These results agree with Gous [25] who reported poor growth rates and feed conversion ratio in 3-week-old broilers fed with diets containing 250 g/kg of faba bean (*Vicia faba* cv. Fiord), which relatively has a high fibre content. Similarly, the inclusion of white lupins (*Lupinus albus*) at 300 g/kg [26] and pigeon pea (*Cajanus Cajan*) at a rate of 400 g/kg [27] decreased growth performance in broiler chickens.

Likewise, the inclusion of CMBM at 99.98 g/kg increased the AR of NDF while negatively affecting the AR of the essential amino acids. These observations could be due to the possible interference of high fibre in the CMBM and trypsin inhibitors and phenolic compounds in the bean [10] which are known to reduce nutrient bioavailability and utilisation. Indeed, the presence of trypsin inhibitors in CMBM [12] forms stoichiometric protease-inhibitor complexes with endogenous proteolytic enzymes and inhibit their action in the gastrointestinal tract [28] either by competitive or allosteric action [29]. Thus, the presence of trypsin and chymotrypsin inhibitors and condensed tannins in marama bean could inhibit digestive enzymes from fully utilising feed components [30]. These findings agreed with Faryadi et al. [31], who reported poor AA digestibility in broiler chickens reared on diets containing 80 g/kg of either soybean hulls or sugar beet pulp.

*Blood parameters*

Haematological and biochemical markers are essential indicators of bird health and have been used in the diagnosis, treatment, and prognosis of numerous illnesses [32]. High dietary fibre level supresses protein and fat digestibility [23], which explains the increase in serum lipase concentration to aid lipid utilisation as dietary CMBM levels increased. The treatments had no influence on serum total protein, albumin, globulin, and albumin-to-globulin ratio, which is not surprising because the diets were isonitrogenous. Aderolu et al. [33] stated that serum protein is a function of protein quality and that a decrease in globulin levels may indicate malnutrition, which suggests that the inclusion of CMBM did not compromise the protein status of the birds. On the other hand, it is rather surprising that neither linear nor quadratic responses were recorded for serum glucose levels, because the high insoluble fibre components in CMBM were expected to slow the substrate passage rate in the gut, thus reducing the digestion and absorption of carbohydrates. Indeed, a highly viscous digesta delay the release of glucose into the bloodstream [34]. Contrastingly, soybean replacement with unconventional protein sources such as lupin (*Lupinus albus*) and lentil (*Lens culinaris*) seeds both at the rate 400 g/kg decreased serum glucose, triglycerides, and cholesterol concentrations in broilers [35,36]. Nevertheless, the reported blood values corresponds with the reference values for healthy quail [1], which suggests that feeding CMBM does not compromise the health status of Jumbo quail. The lack of dietary changes in liver enzymes (GGT and ALKP) further suggests that marama bean could be a safe feed ingredient. In addition, haematological parameters such as neutrophils, monocytes, eosinophils, and basophils, which are white blood cell indicators, increased with dietary MBM levels, suggesting that the immune responses of the birds were not compromised. Contrary to our findings, Emiola et al. [32] noted a decrease in white and red blood cell counts in broilers fed diets with dehulled *Mucuna pruriens var utilis*, which was an indication of compromised immunity. Nonetheless, although feeding diets with CMBM did not compromise the health status of the birds, pre-processing methods (such as decorticating and heating) should be adopted to reduce the negative effects of fibre and trypsin inhibitors on growth performance metrics.

*Carcass, visceral organs, and meat quality*

The functionality and physiology of the gastrointestinal tract in poultry birds are strongly influenced by the amount and type of fibre found in their feeds [37]. Thus, moderate amounts of different fibre components in quail diets could modulate their visceral morphology as well as their growth. Increasing the levels of CMBM in the diet resulted in linear increases in the weights of the proventriculus, gizzard, and small and large intestines. This is attributable to the impact of high dietary fibre in the CMBM group, which could have triggered an anatomical and physiological development of the digestive tract as a strategy to facilitate fibre breakdown [38]. The heavier gizzard weights in Jumbo quail offered the CMBM treatments supports the findings of González-Alvarado et al. [39], who reported that gizzard's muscular activity is stimulated by fibre type and particle size, leading to enlarged gizzards. Likewise, Mateos et al. [37] reported that feeding broilers diets containing 50 g/kg oat hulls or sugar beet resulted in larger gizzards due to extended grinding activities of fibre.

Carcass yield is a crucial economic feature in poultry production, processing, and marketing because it indicates the proportion of live weight converted to marketable meat [40]. Inclusion levels of CMBM reduced slaughter weights and carcass yield of Jumbo quail. These findings corroborate the poor performance results observed in this study, further suggesting that the concentration of trypsin inhibitors and dietary fibre requires prior processing if marama bean is to be used successfully as a nutrient source. Similarly, Diaz et al. [41] reported a decline in carcass weights of broiler chickens reared on diets containing 350 g/kg raw field pea (*Pisum sativum* L.). On one hand, trypsin inhibitors in oilseeds like marama bean increases metabolic activities in internal organs such as the pancreas, resulting in enormous energy usage, which should otherwise be used for muscular development and growth [42]. On the other hand, NSP in CMBM increase digesta viscosity which prohibits enzyme diffusion resulting in poor digestibility and nutrient absorption that are needed for muscle growth [43]. This signifies that pre-processing (e.g., decorticating and heating) of marama bean could be desirable to maximise the amount of saleable or edible meat.

Meat colour at retail shelves or point of purchase is one of the most important aspects that influence the buyer's decision. Consumers tend to favour meat products that have a familiar colour [44]. In this study, feeding diets with varying levels of CMBM did not alter breast meat colour and pH, indicating that the inclusion of CMBM does not alter the appearance of the meat. The lack of alteration in breast meat colour from Jumbo quail reared in CMBM-containing diets could be linked to low levels of carotene pigment in marama, which a lipid-soluble antioxidants responsible for bright red, yellow and orange of plant tissues [45]. The linear decrease in drip loss indicates low glycogen levels in the CMBM groups, since drip loss is strongly influenced by glycolytic potential, pH, and muscle fibre composition [46]. High drip loss in fresh meat indicates poor quality meat characterized by losses in nutrients, flavour and juiciness, which could result in financial losses for the producer [47]. Thus, the increase in drip loss could signify a potential loss of meat nutrients, translating to the production of tough and poor quality [48] quail meat.

## Conclusion

The inclusion of corticated marama bean meal up to 99.98 g/kg in Jumbo quail diets compromised growth performance parameters, physiological responses, slaughter weights and carcass production. Likewise, feeding diets with corticated marama bean meal exacerbated overall mortality and breast meat drip loss. Thus, decortication and thermal treatment of the beans prior to inclusion in diets of Jumbo quail is recommended to mitigate the antinutritional activities of fibre in marama coat and trypsin inhibitors in marama bean, which would allow for successful utilisation.

## CRediT authorship contribution statement

**Mveleli Marareni:** Data curation, Software, Investigation, Writing – original draft. **Elijah G. Kiarie:** Validation, Visualization, Writing – review & editing. **Caven M. Mnisi:** Conceptualization, Project administration, Methodology, Resources, Supervision, Software, Formal analysis, Validation, Visualization, Writing – original draft, Writing – review & editing.

## Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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