

## Evaluation of body calcium and phosphorus composition of the Cobb 700 genotype during growth

F. Salisbury<sup>a</sup>, R. M. Gous<sup>b</sup>, A. J. Cowieson<sup>c</sup>, C. Jansen Van Rensburg<sup>a</sup>, S. J. Rochell<sup>d</sup> and H. Walters<sup>e</sup>

<sup>a</sup>Department of Animal Science, University of Pretoria, Pretoria, South Africa; <sup>b</sup>School of Agricultural, Earth and Environmental Sciences, University of KwaZulu-Natal, Pietermaritzburg, South Africa; <sup>c</sup>Department of Animal Nutrition and Health, DSM-Firmenich Nutritional Products, Kaiseraugst, Switzerland; <sup>d</sup>Department of Poultry Science, Auburn University, Auburn, AL, USA; <sup>e</sup>Tyson Foods, Springdale, AR, USA

### ABSTRACT

1. The amount of calcium and phosphorus in the empty, feather-free bodies of Cobb 700 breed males and females was measured from hatch to 15 weeks of age.  
2. A four-phase *ad libitum* feeding programme was used to feed 400 chicks of each sex. Feeds contained commercial levels of calcium and phosphorus which met or exceeded the requirements for energy, protein and amino acids. All birds were weighed weekly. Ten birds per sex were sampled at 0, 7, 14, 28, 42, 56, 70, 84 and 105 d of age. Defeathered birds were minced, freeze-dried and then analysed for protein, ash, calcium and phosphorus content.  
3. Amounts of Ca in males and females at 105 d averaged 54 and 37 g, respectively, and 105 d P content in males and females averaged 35 and 27 g, respectively. The allometry of ash, Ca and P with empty, feather-free body protein was assessed. Ash was isometric with body protein, while an allometric exponent greater than 1 was established for Ca and P with body protein.  
4. The allometric models and means for Ca and P relative to body protein were compared with models and means found in the literature. Comparison of the Ca/protein ratios with previous data suggested that modern broilers may not reach their potential bone mineral deposition with current dietary guidelines, even when growth rates were maximised.  
5. Theoretical allometric relationships between Ca and P in bone and bone-free body and body protein have now been proposed. The allometric relationship between Ca and P and empty, feather-free body protein offers a suitable way to model the growth of these minerals in the broiler body.

### ARTICLE HISTORY

Received 26 March 2024  
Accepted 4 July 2024

### KEYWORDS

Calcium; phosphorus; modelling; growth; broilers



## Introduction

Recommendations for calcium (Ca) and phosphorus (P) in broiler feeds may be used in least-cost feed formulation for birds at different ages. However, these have limited use to the nutritionist who seeks to maximise financial performance, limit environmental impacts and promote broiler welfare. Different feeding strategies are needed as economic conditions and production objectives change. Furthermore, interactions between Ca and P, as well as additives such as phytase, may affect the ability of birds to use the minerals present in feed (Reis et al. 2023). Diet composition affects the efficacy of phytase. For example, Ca levels in feed will affect the amount of available P released (Amerah et al. 2014; Sommerfeld et al. 2018) and increasing levels of non-phytate P (nPP) may reduce phytate P (PP) degradation (Shastak et al. 2014). The following paper proposes that nutritionists use an alternative approach that predicts broiler responses to Ca and P and is integrated with an established broiler growth model (EFG Software 2019).

Modelling broiler nutrition applies the factorial method (WPSA 1985). This is an approach that seeks first to establish the amounts of the nutrient that are required for potential growth and maintenance (Salisbury et al. 2021). Once these are known, the capacity of the formulated feed to meet requirements can be assessed. This requires a prediction of feed intake, which should be based on the theory regarding how the animal attempts to reach its ideal state within the constraints imposed by its environment and feed (Emmans

1981). Furthermore, an understanding of the availability of the Ca and P in the feed is needed. This may require a model of minerals available from a complete feed, rather than the combination of digestible minerals from individual feed components (Reis et al. 2023). The diet may then be optimised to achieve the production objectives (Gous and Berhe 2006).

The growth of protein, fat and water in the broiler's body is well understood and can be modelled accurately when feed composition is known (Emmans 1981; Emmans and Fisher 1986; Gous et al. 1999). However, this is not the case for Ca and P. The composition of body ash during growth has seldom been reported for broilers (Caldas et al. 2019; Dieckmann 2004; Hurwitz and Plavnik 1986). The Gompertz equation is appropriate for describing the potential growth of each chemical component from hatching to maturity (Emmans 2022), *i.e.* when conditions (feed, environment, health) do not limit performance. Feather-free-body protein (FFBP), water, fat and ash share a common rate of maturing with empty, feather-free body (EFFB). Hence, it has been established that the chemical components of EFFB can be calculated from body protein, using allometry (Emmans 2022). If the allometric exponent (*b*) is 1, then an isometric relationship exists between the two variables (*i.e.* directly proportional to one another). If  $b > 1$ , the ratio between W and FFBP will increase over time and if  $b < 1$ , this ratio will decrease.

**CONTACT** R. M. Gous  [gous@ukzn.ac.za](mailto:gous@ukzn.ac.za)  School of Agricultural, Earth and Environmental Sciences, University of KwaZulu-Natal, P/Bag X01, Scottsville 3209, South Africa

© 2024 The Author(s). Published by Informa UK Limited, trading as Taylor & Francis Group.

This is an Open Access article distributed under the terms of the Creative Commons Attribution-NonCommercial-NoDerivatives License (<http://creativecommons.org/licenses/by-nc-nd/4.0/>), which permits non-commercial re-use, distribution, and reproduction in any medium, provided the original work is properly cited, and is not altered, transformed, or built upon in any way. The terms on which this article has been published allow the posting of the Accepted Manuscript in a repository by the author(s) or with their consent.

While ash has been shown to be isometric with FFBP under ideal conditions (Gous et al. 1999), the composition of EFFB ash during growth has seldom been reported for broilers (Dieckmann 2004; Hurwitz and Plavnik 1986). The changes in body Ca and P must be established from hatch to a body weight close to maturity if the Gompertz parameters are to be estimated and allometric relationships tested. Caldas et al. (2019) reported BW (including feathers, after 6 h of feed deprivation), water, body protein, fat, ash, Ca and P up to 60 d of age. Gompertz curves were fitted to the data, but allometry between chemical components was not investigated. The rates of maturing in terms of protein, Ca and P were the same in this study, suggesting that allometry might be established. This appeared to be the longest duration study published since Hurwitz and Plavnik (1986), which ran for 70 d of age. Neither of these approached full maturity in broilers.

If birds are to be fed precisely to meet their requirements, it is necessary to measure the potential accumulation of Ca and P in the broiler body. Relationships between feather-free body protein, which have been established for other chemical and physical components (Gous et al. 2019b), allow the potential growth of Ca and P in bone and soft tissue to be modelled directly. The possible allocation of Ca and P into bone and soft tissue as subsets of total body content have yet to be explored.

The following paper describes a study in which Ca and P allocation were measured. Ash samples used in the study were from a more comprehensive evaluation of the Cobb 700 genotype, described in Gous et al. (2024). Unlike other strains currently used in commercial production, this strain has not been previously assessed to establish its potential growth parameters.

## Materials and methods

One-day-old male and female Cobb 700 broiler chicks were housed in floor pens in an environmentally controlled room, with fresh pine shavings added to used litter as bedding. Each pen was provided with an appropriate number of nipple drinkers and hanging feeders with a commercial-style feed pan. Upon arrival, the entire population of 1000 chicks was group weighed and counted to obtain a mean weight for each sex. Twenty chicks of each sex were then randomly selected and weighed as a group to confirm that their weight fell within 2% of the calculated amount based on the population mean. A total of 400 chicks were selected for each sex (800 birds in total) and were assigned randomly to one of 20 replicate pens. Each pen measured  $1.22 \times 1.52$  m, providing  $0.093 \text{ m}^2$  per bird. The trial continued for 15 weeks. Broilers had free access to water and feed. Light was continuous for the first 24 h after placement, and a 16 h daylength was used for the remainder of the trial (16 L:8D) as this has been shown to reduce mortality, produce stronger bones, improve FCR, whilst enabling broilers to grow as rapidly as those on longer photoperiods (Lewis et al. 2009a, 2009b). A four-phase feeding programme was used (1 to 14, 15 to 28, 29 to 35 and 36 to 112 d of age). The experimental feeds were formulated to contain a constant amount of energy (13.0 MJ AMEn/kg) and meet or exceed the nutritional requirements suggested by the EFG broiler growth model (EFG Software 2019) for males with a potential mature body protein weight

of 1.6 kg and a rate of maturing of 0.038/d. The composition of the experimental feeds is given in Table 1.

After weekly weighings, 10 birds per sex were selected from randomly predetermined pens to be individually weighed and used for processing and body composition analysis on all weeks except weeks 9, 11, 13, and 14. The selection process included five pens from each sex, with two birds being randomly selected from each pen to give a total of 10 birds per sex for each sampling period. The groups used for sampling was alternated throughout the trial to minimise effects of stocking density. Selected birds were moved into separate pens and feed was withdrawn approximately 8 h before processing the following day. Sampled birds, at 1, 7, 14, 21, 28, 35, 42, 56, 70, 84 and 105 d of age, were weighed just prior to slaughter and again after bleeding. All feathers were removed, after which feather-free body weight and the total weight of feathers was measured.

Each bird, without feathers, was ground using a meat grinder (#32 Big Bite, LEM, Chester, OH, U.S.A.). The samples were passed through the grinder three times to ensure adequate size reduction and mixing, after which a 100 g sub-sample of the wet mince was kept for chemical analysis. This was analysed for water (by freeze-drying), protein (Dumas method on a LECO N analyser; AOAC 2003), ash (AOAC 2003), body fat (Soxhlett extraction, AOAC 2003) and gross energy (GE; using an adiabatic bomb calorimeter). Lyophilised samples were digested with nitric acid and total P and Ca concentrations were determined using an inductively coupled plasma mass spectrometer (procedures adapted from AOAC 985.01a,b,d).

Mean weights of the EFFB and its chemical components were calculated by ANOVA using Genstat 23rd edition (VSN International 2022). The mature mass ( $W_m$ ) was adjusted such that the rate of maturing ( $B$ ) parameters was the same as that determined for the EFFB (Gous et al. 2024). This permitted examination of the allometry between chemical components. The adjustment described in Gous et al. (2024) required an iterative process, accomplished by plotting  $G_t$  against age and manipulating the mature weight of each component until the desired  $B$  value was achieved.

The sum of the adjusted mature weights of protein, ash, water and lipid should be consistent with the fitted mature weight of the EFFB, to establish the hypothesis that all components shared a common rate of maturing was valid. This was shown in Gous et al. (2024) for the data in this study. Once the mature weights were adjusted, the values for EFFBP, ash, Ca and P were calculated using the Gompertz equation with the fitted values for  $W_0$ , adjusted  $W_m$  values and  $B$  values for EFFB. Linear regressions for the natural logarithms ( $\ln$ ) of ash, Ca and P against FFBP were fitted. The slopes of these represent the growth coefficient  $b$  (allometric exponent) while the intercepts, exponentiated, represent  $c$  in the allometric equation:

$$W_t = c * FFBP_t^b$$

where  $W_t$  is the weight of a component at time  $t$  (Moughan et al. 1990). Hence an alternative form of the allometric equation was:

$$\ln(W_t) = a + b * \ln(FFBP_t)$$

where  $a = \ln(c)$ .

The suitability of the allometric relationship for modelling bone Ca and P and bone-free body (BFB) Ca and P was investigated. While Ca and P were expected to be found in constant

**Table 1.** Composition (g/kg) of feeds used in the trial.

Ingredient	Phase 1	Phase 2	Phase 3	Phase 4
Yellow maize	566	631	680	747
Soybean oilcake	348	299	260	213
Fat, poultry	41.7	29.5	21.0	7.50
Dicalcium Phosphate	12.0	11.2	11.4	10.6
Limestone	9.84	9.67	9.85	9.67
L-Lysine HCl	5.08	4.14	3.48	1.78
Sodium Bicarbonate	3.02	2.69	2.46	1.60
Salt	2.78	3.04	3.22	3.89
L-Arginine	2.66	1.91	1.39	0.00
Methionine, DL	2.25	1.75	1.50	0.67
Threonine 98%	2.10	1.82	1.61	0.67
Choline Chloride	1.50	1.67	1.79	1.00
L-Valine	0.91	0.88	0.64	0.00
OptiPhos (2000)	0.50	0.50	0.50	0.50
Vitamin & mineral premix <sup>2</sup>	0.30	0.30	0.30	0.30
Danisco Xylanase	0.08	0.08	0.08	0.08
Nutrient content (calculated)				
AMEn (MJ/kg)	13.0	13.0	13.0	13.0
Crude Protein	221	201	185	165
Calcium <sup>1</sup>	8.1	7.7	7.7	7.3
Total phosphorus <sup>1</sup>	5.8	5.5	5.4	5.0
Phytate phosphorus	2.4	2.3	2.3	2.2
Non-phytate phosphorus	3.4	3.2	3.1	2.9
Sodium	2.0	2.0	2.0	2.0
Chloride	3.0	3.0	3.0	3.0
Arginine <sup>3</sup>	15.8	13.7	12.1	9.5
Lysine	15.0	13.0	11.5	9.0
Methionine	5.3	4.6	4.2	3.2
Met + Cys	8.2	7.3	6.8	5.7
Tryptophan	2.5	2.2	2.0	1.8
Threonine	8.9	8.0	7.3	5.8
Isoleucine	8.8	7.9	7.2	6.4
Valine	10.4	9.5	8.6	7.2

<sup>1</sup>The values for Ca and P do not include amounts released by phytase.

<sup>2</sup>Content/kg of diet: Mn = 150.000 mg. Fe = 100.000 mg. Zn = 100.000 mg. Cu = 16.000 mg. I = 1.500 mg. 2Content/kg of product: Folic acid = 1000 mg. Pantothenic acid = 15.000 mg. Niacin = 40.000 mg. Biotin = 60 mg. vit B1 = 1.800 mg. vit B12 = 12.000 mg. vit B2 = 6.000 mg. vit B6 = 2.800 mg. vit D3 = 2.000.000 UI. vit E = 15.000 mg. vit K3 = 1.800 mg. Se = 300 mg.

<sup>3</sup>Digestible amino acid contents given.

concentrations in the BFB, bones may contain varying amounts of ash, and hence Ca and P, depending on the diet (Reis et al. 2023). This required theoretical apportioning of the amounts in the whole body, as the necessary dissections have not been reported in broilers at maturity (approximately 168 d of age). This is required for the establishment of accurate allometric relationships, or even to the end of the growing period. Angel (2007) reported the amounts of Ca and P in the skeleton up to 14 d of age and demonstrated rapid mineralisation bones during this period, but body protein and whole-body minerals were not reported. Coefficients of 0.00031 g BFB Ca/g FFBP and 0.00915 g BFB P/g FFBP were used, and the balance of carcass Ca and P was assigned to bone. These coefficients were derived from a preliminary model based on the study of Caldas Cueva (2015) and briefly described in Salisbury et al. (2021).

Bone Ca:P ratio had a mean value of 2.15 (SE 0.3) which suggested that Ca and P were appropriately assigned to the two pools, BFB and bone. The calculation of means, adjustment for mature values and fitting allometric regressions to the log-transformed data was repeated as described above. The performance of the allometric models was compared with published values.

## Results

The mean weights of the EFFB and components are presented in Table 2. The level of Ca as a proportion of EFFB increased from 0.3% at day old to 0.7% in males and 0.6% in females at 105 d. The level of P as a proportion of EFFB

increased from 0.3% to 0.45% in males and 0.4% in females (Gous et al. 2024). Between 14 and 28 d, ash, Ca and P decreased as a proportion of EFFB before resuming an upward trend.

The fitted Gompertz equation for the growth of EFFB returned a rate of maturing (B) parameter of 0.0417 for males and 0.0402 for females.

The chemical component mature levels were adjusted so that B was uniform, and these values are summarised in Table 3. The means for each weighing point were recalculated using the Gompertz function. All the component means were then log-transformed to allow the allometry between them to be assessed.

The parameters of the allometric equation are summarised in Table 4. There was a significant difference between the intercepts (*a* values) for males and females, but the slopes were the same. The allometric coefficients for the Ca and P distributed to the bone and BFB pools are shown in Table 5.

## Discussion

This was part of a study to establish the potential growth of the chemical components of male and female Cobb 700 broilers. While this type of experiment has been successfully conducted for other broiler genotypes to estimate chemical component growth (Gous et al. 2019a, 2024; Vargas et al. 2020), potential Ca and P deposition have not been assessed. Hence the conditions that would be non-limiting for bone

**Table 2.** Mean weight<sup>1</sup> ± SD (g) of empty, feather-free body (EFFB), feather-free body protein (FFBP), ash, calcium (Ca) and phosphorus (P) at weekly intervals during the growth of female and male Cobb 700 broilers to 105 d of age.

Day	Male										Female									
	EFFB ± SD		FFBP ± SD		Ash ± SD		Ca ± SD		P ± SD		EFFB ± SD		FFBP ± SD		Ash ± SD		Ca ± SD		P ± SD	
0	39	0.34	5	0.27	0.6	0.04	0.1	0.01	0.1	0.01	39	0.25	5	0.47	0.6	0.03	0.1	0.01	0.1	0.01
7	133	3.22	19	1.57	2.8	0.25	0.6	0.05	0.5	0.04	132	4.59	18	1.69	2.7	0.24	0.5	0.05	0.5	0.05
14	385	11.3	67	5.96	9.7	1.10	2.3	0.59	2.0	0.40	379	11.6	69	5.82	9.5	1.06	2.3	0.43	1.9	0.29
21	886	24.6	143	8.84	19.2	2.20	4.1	0.91	3.3	0.44	844	20.6	137	11.8	19.1	2.45	4.7	0.91	3.5	0.60
28	1585	40.8	258	10.2	31.6	6.40	7.7	1.18	5.9	0.61	1470	32.8	222	22.2	30.2	3.86	6.1	1.27	4.6	0.86
35	2424	64.9	391	29.4	56.7	4.70	12.7	1.23	9.0	0.78	2148	42.0	334	40.7	47.4	4.90	10.1	2.72	7.5	1.82
42	3273	80.9	538	38.2	74.1	11.1	17.4	4.26	12.6	2.28	2820	59.5	469	42.6	59.7	9.03	14.0	3.39	10.2	1.59
49	4211	107	664	71.9	98.5	20.8	24.8	7.65	16.5	3.85	3509	78.9	559	61.2	71.9	17.6	15.9	6.81	11.9	2.93
56	5024	156	824	89.3	122	21.2	29.0	8.29	19.8	3.96	4076	98.6	653	48.1	91.3	20.5	21.6	7.09	15.0	3.15
70	5585	205	1018	104	152	31.9	37.7	139	25.9	6.85	4535	107	847	117	108	16.5	28.6	13.8	20.4	7.63
85	6549	185	1204	139	177	25.5	45.0	6.15	31.0	3.42	5093	181	989	106	128	20.9	33.7	15.8	23.4	7.31
105	7012	255	1383	221	167	58.7	53.7	16.6	35.3	8.67	5397	226	1110	144	145	24.0	37.3	9.62	26.6	3.03

<sup>1</sup>10 birds per sex per sampling age.**Table 3.** Gompertz parameters for empty, feather-free body, protein, ash, calcium and phosphorus.

	Males					Females				
	EFFB	CP	Ash	Ca	P	EFFB	CP	Ash	Ca	P
W <sub>m, g</sub>	8380	1562	218	57.1	37.5	6940	1191	198	40.8	28.6
W <sub>0, g</sub>	39	5.1	0.7	0.1	0.1	34	4.6	0.6	0.1	0.1
B <sub>1</sub> /d	0.0417	0.0417	0.0417	0.0417	0.0417	0.0402	0.0402	0.0402	0.0402	0.0402

**Table 4.** Allometric coefficients for ash, calcium and phosphorus relative to feather-free body protein.

Coeff <sup>1</sup>	Males		Females	
	a	b	a	b
Ash	-1.973 (±0.0001)	0.9904 (±0.00004)	-1.957 (±0.0002)	0.9904 (±0.00004)
Ca	-4.074 (±0.0078)	1.111 (±0.0023)	-4.143 (±0.0134)	1.111 (±0.0023)
P	-4.123 (±0.0031)	1.062 (±0.0009)	-4.162 (±0.0054)	1.062 (±0.0009)

<sup>1</sup>The coefficients are those for the equation  $\ln(W_t) = a + b * \ln(FFBP_t)$  where  $W_t$  is the weight of ash, Ca or P at time  $t$  and  $FFBP_t$  is the weight of protein in the empty, feather-free body at time.**Table 5.** Allometric coefficients for the Ca and P distributed to the bone and bone-free body pools.

Coeff <sup>1</sup>	a		
	Males	Females	b
Ca BFB	-7.176 (±0.0061)	-7.124 (±0.0106)	1.106 (±0.0018)
Ca bone		-4.234 (±0.0219)	1.124 (±0.0039)
P BFB	-5.130 (±0.0065)	-5.087 (±0.0712)	1.055 (±0.0019)
P bone		-4.748 (±0.0214)	1.083 (±0.0038)

<sup>1</sup>The coefficients are those for the equation  $\ln(W_t) = a + b * \ln(FFBP_t)$  where  $W_t$  is the weight of ash, Ca or P at time  $t$  and  $FFBP_t$  is the weight of protein in the empty, feather-free body at time.

growth were not known *a priori* and birds were fed commercially accepted levels of Ca and P.

It has been proposed that Ca and P are isometric with empty body protein when pigs are fed to maximum (potential) protein growth and with an adequate supply of Ca and P in the diet (Hendriks and Moughan 1993; Misiura et al. 2020; Pettey et al. 2015). Hurwitz et al. (1987) fitted a logarithmic function to the proportion of Ca in the EFFB over time, which suggested that this increased in the first three weeks before levelling off. Similarly, Angel (2007) found that the skeleton was most rapidly mineralised in the first few weeks of age.

Létourneau-Montminy et al. (2015) proposed that soft tissue and bone should be modelled separately in pigs. Further development of this model has been reported (Lautrou et al. 2020). Recent models have applied this segregation to Ca and P in broilers (Reis et al. 2023; Salisbury et al. 2021) and in pigs (Misiura et al. 2020). Reis et al. (2023)

assumed that Ca in bone is a constant proportion (22%) of body ash. Ash was calculated from an allometric relationship with body protein, derived from Hruby et al. (1994). The allometric coefficient of 0.898 g/g would result in a slight decline in the ratio between bone Ca and body protein over the growth period in this model. The small amount of Ca in the soft tissue is unlikely to contribute to a significant change in whole-body Ca as a proportion of ash. Although bone P was calculated as half of bone Ca, the separation of P into bone and soft tissue portions, and the calculation of P in the latter in proportion to the growth of lipid, feathers and body protein may result in whole body P varying as a proportion of ash in this model.

In the present study, the relationship between body ash and FFBP has a coefficient (exponent) that was not significantly different from 1. Hence an isometric relationship between these components was suggested. Ash can be calculated as  $0.14 \times$  body protein. This isometry contrasted with the relationship

between Ca and P and protein, for which the ratio with FFBP was higher ( $b > 1$ ). These relationships suggested that ash composition changes as the bird grows. The larger constant terms in the allometric equations for Ca and P suggested that bones may be heavier (more mineralised) in male birds in relation to body protein, but the common slopes indicated that the relationships with protein change in a similar way as the bird grows. The proportions of Ca and P in bone ash were close to those in stoichiometric hydroxyapatite ( $\text{Ca}_{10}(\text{PO}_4)_6(\text{OH})_2$ , 39.9% Ca and 18.5% P, Ca/P mass ratio 2.16; Browning and Cowieson 2014). Slightly lower levels of Ca were sometimes observed, resulting in a lower Ca/P ratio (Dersjant-Li et al. 2018; Skinner and Waldroup 1995). While P in soft tissue ash (0.13–0.17 g P/g ash) can be similar to that in bone (0.16–0.18 g P/g ash; Rousseau 2013), the Ca concentration tends to be much lower in soft tissue than in bone (Grey et al. 1983). Hence changes in ash composition may be a result of changing proportions of ash in bone and BFB.

The results suggested that the allometric relationship with FFBP was a feasible way to model Ca and P growth in the broiler. When the observed weights of Ca and P were plotted against the expected values calculated using the allometric models, the fitted equations (for male broilers) were as follows:

$$Ca_{\text{observed}} = 0.210(N.S.) + 0.987(\pm 0.0299) \times Ca_{\text{expected}}$$

$$P_{\text{observed}} = 0.146(N.S.) + 1.006(\pm 0.0387) \times P_{\text{expected}}$$

The means and modelled values from this experiment were compared with data from two previous studies using the ratio between Ca or P and the FFBP over time (Caldas et al. 2019; Dieckmann 2004; Figures 1 and 2).

Dieckmann (2004) fed Ross chicks a range of Ca and P levels whereby the Ca/P ratio in the feed varied. The graphs representing these data represent the average Ca or P deposition (g/kg FFBP) across the treatments. Caldas et al. (2019) fed male Cobb broilers according to the breed guidelines. Dieckmann (2004) showed that the Ca/FFBP and P/FFBP ratios increased initially before levelling off. The plots of the means from the present study and those from Caldas et al. (2019) increased initially but decreased after the first 14 d, before increasing gradually once again. The allometric model ratios were consistently lower than measured values up to 21 d, but the data fitted the model closely from this age until the birds reached 105 d of age. The modelled values for Ca at 42 d (5.5 g/kg BW) corresponded closely with the 5.75 g Ca/kg BW reported by Angel, cited in Reis et al. (2023).

The Ca (g/kg FFBP) values derived from Dieckmann (2004), 44 g Ca/kg FFBP and 32 g P/kg FFBP at 21 d of age) were higher than those in the present study which reached 34 g Ca/kg FFBP and 29 g P/kg FFBP at 14 d of age before decreasing to 28 g Ca/kg FFBP and 23 g P/kg FFBP at 21 d of age. This could reflect a genotype difference between the 2004 Ross and 2023 Cobb breed used. If the potential level of Ca was lower for the newer strain, this implied less mineralisation in the skeleton or a larger proportion of mineral in muscle tissue, where Ca and P were present in smaller amounts. However, the 42 d Ca/P ratio was 1.4 in both studies. A change in proportion between muscle and bone would

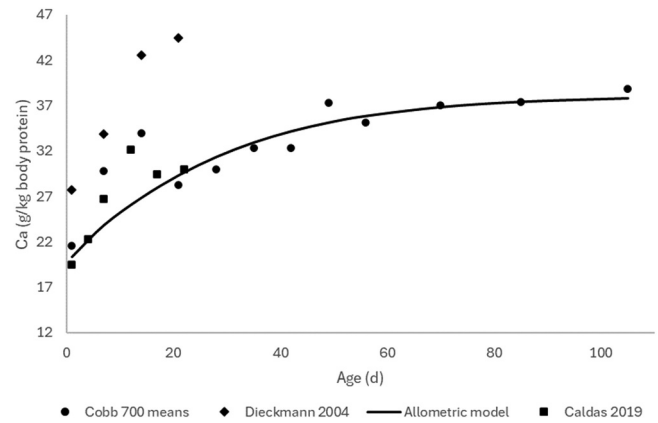


Figure 1. Modelled and experimental values demonstrating the relationship between calcium in the broiler body and feather-free protein weight over the growing period.

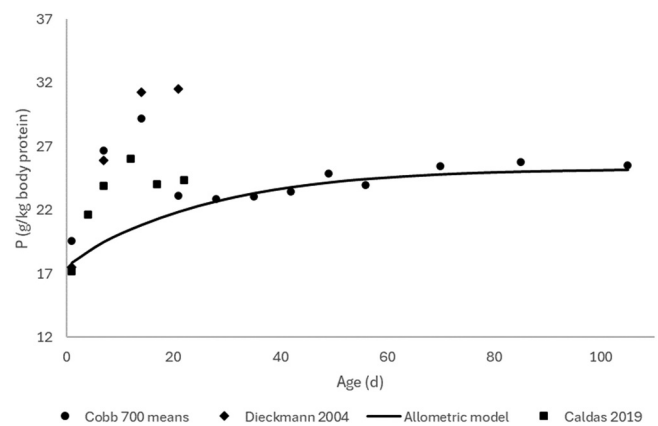
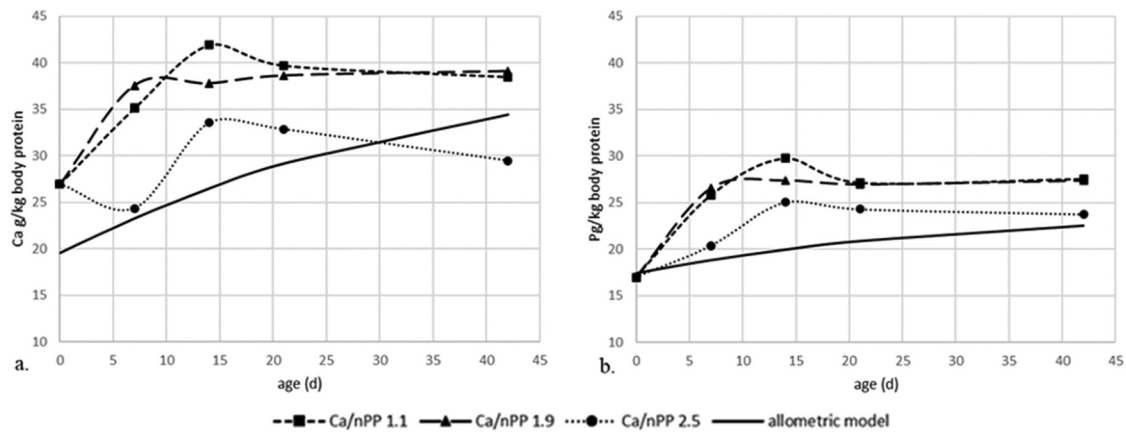


Figure 2. Modelled and experimental values demonstrating the relationship between phosphorus in the broiler body and feather-free protein weight over the growing period.

be expected to alter this ratio, due to the differing proportions of Ca and P in these tissues. If the lower ratio and the decrease in mineral content relative to protein after 14 d was not a result of changing genetic potential, then this study and that of Caldas et al. (2019) suggested that the Cobb birds were not able to reach their potential growth for Ca and P.

It is important to note that an allometric relationship between Ca or P and protein does not imply that the mineral is in some way chemically associated with protein. Allometric relationships are likely to arise from a common impetus provided by the growth of the animal. Hence allometry of bone mineral is calculated with whole body protein, not with bone protein, even though the latter is a substantial component of the bone mass. The division of minerals between bone and soft tissue is, to some extent, a theoretical construct that allows deviation from the allometric relationship when nutrition is limiting. Isometry between body protein and BFB Ca and P can be maintained while bone Ca and P are reduced if these nutrients are limiting. If protein is limiting, bone Ca and P may continue to accumulate while body protein grows more slowly. The division of Ca and P between BFB and bone proposed here suggested that, if BFB Ca and P were isometric with body protein, bone Ca and P have an allometric relationship with body protein and increased in proportion over the growing period.



**Figure 3.** Modelled and experimental values demonstrating the relationship between a. calcium and b. phosphorus and body protein over the growing period with different Ca/P ratios (source: Dieckmann 2004).

**Table 6.** Feed composition (g/kg) in body composition trial (Dieckmann 2004).

	Feed	Ca	Total P	PP	nPP	Ca/nPP	Ca:P
1	Starter	7.94	8.95	1.7	7.25	1.10	0.89
	Grower	7.32	7.45	1.6	5.85	1.25	0.98
2	Starter	7.59	5.69	1.7	3.99	1.90	1.33
	Grower	7.17	4.81	1.6	3.21	2.23	1.49
3	Starter	6.15	4.17	1.7	2.47	2.49	1.47
	Grower	4.67	3.09	1.6	1.49	3.13	1.51

Functions estimating restrictions on growth from bone mineral deficiency may be different from those required when mineral supply is insufficient for soft tissue growth. Hence the changes in allometry reported when the diet was limiting in either P or protein, as reported in pigs ( $b > 1$  for body P allometry with body protein,  $b < 1$  when protein was non-limiting, but P was deficient). If effects on body weight gain are the only concern, segregation may be unnecessary. An empirical function may be applied to whole body Ca and P to reduce BWG when these are deficient. However, if skeletal integrity is a concern, an accurate model of the apportioning of mineral to BFB and bone may be required, but this requires further experiments whereby a consistent dissection protocol is applied. The results of mineral analysis after dissection in two studies showed very different proportions of Ca and P in the skeleton and soft tissue (Angel 2007; Van Krimpen et al. 2013). This highlighted the difficulties inherent in the dissection process. Soft tissue must be carefully separated from bone, which is time consuming and may leave soft tissue adhering to the bone. However, it allows separation without physical or chemical changes to both portions of the body. Boiling may alter the composition of the soft tissue portion, as will enzymatic cleaning of bone. Furthermore, a decision must be made regarding whether cartilage is included in the skeleton portion of the body. Leaving the cartilage caps on the tibia before analysis may reduce the ash content by as much as 12% (Fritz and Halloran 1943). This may explain the low ash content of the skeleton in the study reported by Angel (2007). Van Krimpen et al. (2013) found that only approximately 75% of the body calcium was in the skeleton, which suggested that the feet, head and beak were included in the non-skeleton portion. These accounted for a decreasing proportion of the calcium in the skeleton, from 26% at 7 days to 20% at 21 d (Itoh and Hatano 1964).

Until the interactions between Ca and P have been successfully modelled, trials to assess the potential growth of

these minerals in broilers may be more successful when a variety of Ca and P levels, as well as different Ca:P ratios, are fed, as they were in the study of Dieckmann (2004; Figure 3, Table 6).

The feeds which produced the highest ratios between Ca and FFBP contained lower Ca/P ratios in feed than are normally fed (Angel et al. 2015). In the third feed used, Ca and P were fed at lower levels with a wider Ca/P ratio. The Ca and P deposition in bone appeared to be compromised, but there were no significant differences in body weight gain.

## Conclusions

It is most likely that Cobb 700 broilers in the present study were not fed to reach their potential deposition of Ca and P. However, direct allometric relationships between these minerals and body protein appeared to show promise for modelling potential growth of Ca and P. This can provide a target for producers who seek to optimise skeletal development. Once bone mineralisation can be predicted, available minerals in the diet can be calculated and the effects of deficiency on BWG can be modelled. This will make it possible to balance the objectives of increased BWG, adequate skeletal integrity and minimise P excretion to optimise broiler production for the individual producer.

## Disclosure statement

No potential conflict of interest was reported by the author(s).

## ORCID

F. Salisbury  <http://orcid.org/0000-0001-5429-6167>  
 R. M. Gous  <http://orcid.org/0000-0002-5457-3643>  
 S. J. Rochell  <http://orcid.org/0000-0003-0839-9389>

## References

- AMERAH, A. M., P. W. PLUMSTEAD, L. P. BARNARD, and A. KUMAR. 2014. "Effect of Calcium Level and Phytase Addition on Ileal Phytate Degradation and Amino Acid Digestibility of Broilers Fed Corn-Based Diets." *Poultry Science* 93 (4): 906–915. <https://doi.org/10.3382/ps.2013-03465>.
- ANGEL, R. 2007. "Metabolic Disorders: Limitations to Growth of and Mineral Deposition into the Broiler Skeleton After Hatch and Potential Implications for Leg Problems." *Journal of Applied Poultry Research* 16 (1): 138–149. <https://doi.org/10.1093/japr/16.1.138>.
- ANGEL, R., E. JIMÉNEZ-MORENO, W. LI, S.-W. KIM, M. PROSZKOWIEC-WEGLARZ, and F. ESCRIBANO. 2015. "Dietary Calcium, Phosphorus and Phytase Interactions." In *Proceedings of the XXXI Curso de Especialización FEDNA*, 111–120.
- AOAC. 2003. *Official Methods of Analysis of the Association of Official Analytical Chemists*. 17th ed. Arlington, VA: AOAC.
- BROWNING, L. C., and A. J. COWIESON. 2014. "Effect of Vitamin D3 and Strontium on Performance, Nutrient Retention, and Bone Mineral Composition in Broiler Chickens." *Animal Production Science* 54 (7): 942–949. <https://doi.org/10.1071/AN13091>.
- CALDAS CUEVA, J. V. 2015. "Calorimetry and Body Composition Research in Broilers and Broiler Breeders." PhD thesis, University of Arkansas.
- CALDAS, J. V., N. BOONSINCHAI, J. R. WANG, J. A. ENGLAND, and C. N. COON. 2019. "The Dynamics of Body Composition and Body Energy Content in Broilers." *Poultry Science* 98 (2): 866–877. <https://doi.org/10.3382/ps/pey422>.
- DESIJANT-LI, Y., C. EVANS, and A. KUMAR. 2018. "Effect of Phytase Dose and Reduction in Dietary Calcium on Performance, Nutrient Digestibility, Bone Ash and Mineralization in Broilers Fed Corn-Soybean Meal-Based Diets with Reduced Nutrient Density." *Animal Feed Science and Technology* 242:95–110. <https://doi.org/10.1016/j.anifeeds.2018.05.013>.
- DIECKMANN, A. 2004. "Beiträge zur Optimierung der Phosphorversorgung von wachsenden Broilern." PhD thesis, Martin-Luther-Universität.
- Es Software. 2019. "EFG Software". Accessed May 1, 2023. <https://www.efgsoftware.net>.
- EMMANS, G., and C. FISHER. 1986. "Problems in Nutritional Theory." In *Nutrient Requirements of Poultry and Nutritional Research*, edited by C. Fisher and K. N. Boorman, 9–39. London, UK: Butterworths.
- EMMANS, G. C. 1981. "A Model of the Growth and Feed Intake of Ad Libitum Fed Animals, Particularly Poultry." In *Computers in Animal Production*, edited by G. M. Hillyer, C. T. Whittemore, and R. G. Gunn, 103–110. Cambridge, UK: British Society of Animal Production.
- EMMANS, G. C. 2022. "The Potential Post-Hatching Growth of Domestic Birds is Sufficiently Described by the Gompertz Function." *British Poultry Science* 63 (5): 701–719. <https://doi.org/10.1080/00071668.2022.2083941>.
- FRITZ, J. C., and H. R. HALLORAN. 1943. "Miscellaneous Observations on the A.O.A.C. Vitamin D Assay." *Poultry Science* 22 (4): 314–322. <https://doi.org/10.3382/ps.0220314>.
- GOUS, R., E. MORAN, H. STILBORN, G. BRADFORD, and G. EMMANS. 1999. "Evaluation of the Parameters Needed to Describe the Overall Growth, the Chemical Growth, and the Growth of Feathers and Breast Muscles of Broilers." *Poultry Science* 78 (6): 812–821. <https://doi.org/10.1093/ps/78.6.812>.
- GOUS, R. M., and E. T. BERHE. 2006. "Modelling Populations for Purposes of Optimization." In *Mechanistic Modelling in Pig and Poultry Production*, edited by R. Gous, C. Fisher, and T. Morris, 76–96. Wallingford, UK: CABI. <https://doi.org/10.1079/9781845930707.0000>.
- GOUS, R. M., C. FISHER, E. TŮMOVÁ, V. MACHANDER, D. CHODOVÁ, J. VLČKOVÁ, L. UHLÍŘOVÁ, and M. KETTA. 2019a. "The Growth of Turkeys 1. Growth of the Body and Feathers and the Chemical Composition of Growth." *British Poultry Science* 60 (5): 539–547. <https://doi.org/10.1080/00071668.2019.1622076>.
- GOUS, R. M., C. FISHER, E. TŮMOVÁ, V. MACHANDER, D. CHODOVÁ, J. VLČKOVÁ, L. UHLÍŘOVÁ, and M. KETTA. 2019b. "The Growth of Turkeys. 2. Body Components and Allometric Relationships." *British Poultry Science* 60 (5): 548–553. <https://doi.org/10.1080/00071668.2019.1622077>.
- GOUS, R. M., H. WALTERS, S. J. ROCHELLE, and G. C. EMMANS. 2024. "Evaluation of the Potential Growth and Body Composition of the Cobb 700 Genotype." *British Poultry Science*. <https://doi.org/10.1080/00071668.2024.2328680>.
- GREY, T. C., D. ROBINSON, J. M. JONES, S. W. STOCK, and N. L. THOMAS. 1983. "Effect of Age and Sex on the Composition of Muscle and Skin from a Commercial Broiler Strain." *British Poultry Science* 24 (2): 219–231. <https://doi.org/10.1080/00071668308416733>.
- HENDRIKS, W. H., and P. J. MOUGHAN. 1993. "Whole-Body Mineral Composition of Entire Male and Female Pigs Depositing Protein at Maximal Rates." *Livestock Production Science* 33 (1): 161–170. [https://doi.org/10.1016/0301-6226\(93\)90247-F](https://doi.org/10.1016/0301-6226(93)90247-F).
- HRUBY, M., M. L. HAMRE, and C. N. COON. 1994. "Growth Modelling as a Tool for Predicting Amino Acid Requirements of Broilers." *Journal of Applied Poultry Research* 3 (4): 403–415. <https://doi.org/10.1093/japr/3.4.403>.
- HURWITZ, S., S. FISHMAN, and H. TALPAZ. 1987. "Model of Plasma Calcium Regulation: System Oscillations Induced by Growth." *American Journal of Physiology-Regulatory, Integrative and Comparative Physiology* 252 (6): R1173–R1181. <https://doi.org/10.1152/ajpregu.1987.252.6.R1173>.
- HURWITZ, S., and I. PAVNIK. 1986. "Carcass Minerals in Chickens (*Gallus domesticus*) During Growth." *Comparative Biochemistry and Physiology* 83A (2): 225–227. [https://doi.org/10.1016/0300-9629\(86\)90565-7](https://doi.org/10.1016/0300-9629(86)90565-7).
- Itoh, H., and T. HATANO. 1964. "Comparison of Calcium Metabolism in Various Bones of Growing Chicks in Varying States of Vitamin D Supplementation." *Poultry Science* 43 (1): 70–76. <https://doi.org/10.3382/ps.0430070>.
- LAUTROU, M., C. POMAR, J. Y. DOURMAD, A. NARCY, P. SCHMIDELY, and M. P. LÉTOURNEAU-MONTMINY. 2020. "Phosphorus and Calcium Requirements for Bone Mineralisation of Growing Pigs Predicted by Mechanistic Modelling." *Animal* 1–10. <https://doi.org/10.1017/S1751731120001627>.
- LÉTOURNEAU-MONTMINY, M. P., A. NARCY, J. Y. DOURMAD, T. D. CRENSHAW, and C. POMAR. 2015. "Modelling the Metabolic Fate of Phosphorus and Calcium and the Dynamics of Body Ash Content in Growing Pigs." *Journal of Animal Science* 93 (3): 1200–1217. <https://doi.org/10.2527/jas2014-8519>.
- LEWIS, P. D., R. DANISMAN, and R. GOUS. 2009a. "Photoperiodic Responses of Broilers. I. Growth, Feeding Behaviour, Breast Meat Yield, and Testicular Growth." *British Poultry Science* 50 (6): 657–666. <https://doi.org/10.1080/00071660903338452>.
- LEWIS, P. D., R. DANISMAN, and R. M. GOUS. 2009b. "Photoperiodic Responses of Broilers. III. Tibial Breaking Strength and Ash Content." *British Poultry Science* 50 (6): 673–679. <https://doi.org/10.1080/00071660903365612>.
- MISIURA, M. M., J. A. N. FILIPE, C. L. WALK, and I. KYRIAZAKIS. 2020. "How Do Pigs Deal with Dietary Phosphorus Deficiency?" *The British Journal of Nutrition* 124 (3): 256–272. <https://doi.org/10.1017/S0007114520000975>.
- MOUGHAN, P., W. SMITH, and E. STEVENS. 1990. "Allometric Growth of Chemical Body Components and Several Organs in the Pig (20–90 Kg Liveweight)." *New Zealand Journal of Agricultural Research* 33 (1): 77–84. <https://doi.org/10.1080/00288233.1990.10430663>.
- PETTEY, L. A., G. L. CROMWELL, Y. D. JANG, and M. D. LINDEMANN. 2015. "Estimation of Calcium and Phosphorus Content in Growing and Finishing Pigs: Whole Empty Body Components and Relative Accretion Rates." *Journal of Animal Science* 93 (1): 158–167. <https://doi.org/10.2527/jas.2014-7602>.
- REIS, M. P., C. COUTURE, N. K. SAKOMURA, L. HAUSCHILD, C. R. ANGEL, A. NARCY, and M. P. LÉTOURNEAU-MONTMINY. 2023. "A First Model of the Fate of Dietary Calcium and Phosphorus in Broiler Chickens." *Animal* 100896. <https://doi.org/10.1016/j.animal.2023.100896>.
- ROUSSEAU, X. R. 2013. "Optimisation de l'utilisation de phosphore alimentaire chez le porc et le poulet en croissance." PhD thesis, Université François-Rabelais de Tours.
- SALISBURY, F., A. COWIESON, and R. GOUS. 2021. "Constraints on the Modelling of Calcium and Phosphorus Growth of Broilers:

- A Systematic Review.” *World’s Poultry Science Journal* 77 (4): 775–795. <https://doi.org/10.1080/00439339.2021.1974804>.
- SHASTAK, Y., E. ZELLER, M. WITZIG, M. SCHOLLENBERGER, and M. RODEHUTSCORD. 2014. “Effects of the Composition of the Basal Diet on the Evaluation of Mineral Phosphorus Sources and Interactions with Phytate Hydrolysis in Broilers.” *Poultry Science* 93 (10): 2548–2559. <https://doi.org/10.3382/ps.2014-03961>.
- SKINNER, J. T., and P. W. WALDROUP. 1995. “Allometric Bone Development in Floor-Reared Broilers.” *Journal of Applied Poultry Research* 4 (3): 265–270. <https://doi.org/10.1093/japr/4.3.265>.
- SOMMERFELD, V., M. SCHOLLENBERGER, I. KÜHN, and M. RODEHUTSCORD. 2018. “Interactive Effects of Phosphorus, Calcium, and Phytase Supplements on Products of Phytate Degradation in the Digestive Tract of Broiler Chickens.” *Poultry Science* 97 (4): 1177–1188. <https://doi.org/10.3382/ps/pex404>.
- VAN KRIMPEN, M. M., J. T. M. VAN DIEPEN, P. G. VAN WIKSELAAR, P. BIKKER, and A. W. JONGBLOED. 2013. *Effects of Available Phosphorus (aP), Calcium/aP Ratio, and Growth Rate on P Deposition, P Digestibility, Performance and Leg Quality in Broilers*. Wageningen, Netherlands: Wageningen UR Livestock Research.
- VARGAS, L., N. K. SAKOMURA, B. B. LEME, F. ANTAYHUA, M. REIS, R. GOUS, and C. FISHER. 2020. “A Description of the Potential Growth and Body Composition of Two Commercial Broiler Strains.” *British Poultry Science* 61 (3): 266–273. <https://doi.org/10.1080/00071668.2020.1716300>.
- VSN International. 2022. *GenStat Release 18.1*. Oxford, UK: Clarendon Press.
- WPSA. 1985. “Mineral Requirements for Poultry – Mineral Requirements and Recommendations for Growing Birds.” *World’s Poultry Science Journal* 41 (3): 252–258. <https://doi.org/10.1079/WPS19850021>.