Body Composition Analysis of Chickens by Dual Energy X-Ray Absorptiometry[1]

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ABSTRACT Dual-energy x-ray absorptiometry (DXA) was evaluated as a method for measuring the body composition of growing broiler chickens. A total of 130 chickens, ranging in weight from 400 to 3,290 g, were scanned using a DXA instrument (Lunar™ DPX-L). Single whole-body scans were acquired and analyzed using pediatric total body research software (neonatal mode) or small animal total body research software (detail or high resolution mode). The DXA measurements provided readings of total tissue mass, percentage fat, fat tissue mass, lean tissue mass, and bone mineral content. After scanning, the bodies of the chickens were frozen, then, after removing the feathers, homogenized for chemical determination of fat, water, and protein content. By chemical analysis, the whole body fat content of the chickens ranged from 2.8 to 27.2%, giving rise to DXA R values (ratio of attenuation coefficients) ranging from 1.415 to 1.339. The accuracy of DXA for measuring total body fat was a function of the scanning program and mode and also the size of the bird. The best agreement between DXA and chemical measurements of percentage body fat were obtained when chickens weighing more than 2,000 g were scanned using either the small animal-detail mode or neonatal mode. None of the scan modes proved to be accurate for measuring the fat content of birds weighing less than 2,000 g. The DXA measurement of lean mass of chickens was found to be highly correlated with both total body protein (R^2 = 0.90) and total body water (R^2 = 0.93), but was of little value for predicting percentage values for either. The ratio of DXA bone mineral content to total body ash was 0.77; however, the correlation (R^2) between the two was only 0.46. These results suggest that although the DXA technique is potentially useful for measuring body composition of chickens, considerable refinement is needed prior to routine application.

(*Key words*: chicken, body composition, dual-energy x-ray absorptiometry)

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INTRODUCTION

The concern over increasing levels of fat in broiler chickens accentuates the need for an accurate method for measuring the body composition, especially fat content, of live chickens. A review by Topel and Kauffman (1988) reported on more than 30 techniques for estimating live animal or carcass composition. In recent years, research in the area of body composition analysis has focused on a variety of instruments designed to obtain more detailed information from "inside" the live animal.

Dual-energy x-ray absorptiometry (DXA) evolved from a similar technique known as dual-energy photon absorptiometry or DPA. DPA was originally developed for the measurement of mineral mass and bone density in humans (Peppler and Mazess, 1981). Because of the low radiation dose and the ability to detect the differential attenuation of the radiation by bone, fat, and lean tissue, both of these dual-energy projection methods have received considerable attention for the measurement of human body composition (Lukaski, 1993). DXA is now considered by many as the method of choice for measurement of human body composition. Most DXA measurements of animals have been limited to laboratory animals or the use of pigs for validation of human measurements (Brunton *et al*., 1993; Svendsen *et al*., 1993). There have been no reported attempts to use DXA for measuring body composition of chickens.

The present study was conducted to provide a more extensive evaluation of the use of DXA for the measurement of body composition of chickens. DXA measurements of total fat and lean body mass in chickens were compared with results obtained by chemical analysis.

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[1]Mention of a trade name does not constitute a guarantee or warranty by the USDA and does not imply its approval to the exclusion of other products that may be suitable.

TABLE 1. Description of experiments in which dual-energy x-ray absorptiometry (DXA) scans were performed on chickens

Group	Live (L) or dead (D)	Number of birds	DXA scan mode	Weight range
				(g)
1A	D	28	Neo-Med[1]	403 to 1,782
1B	D	19	Neo-Med	2,001 to 2,908
2A	D	15	SA-Det[2]	439 to 1,700
2B	D	14	SA-Det	2,017 to 3,289
3A	L	35	SA-Det	628 to 1,992
3B	L	19	SA-Det	2,006 to 3,102
3C	L	105	SA-Det	744 to 3,102
4A	D	35	SA-HR[3]	628 to 1,992
4B	D	19	SA-HR	2,006 to 3,102

[1]Neonatal-Medium (scan version 1.2, analysis version 1.5d).
[2]Small animal-Detail (version 1.0d).
[3]Small animal-High resolution (version 1.0d).

MATERIALS AND METHODS

Body composition was measured with a total-body DXA scanner.[2] The basic theory and methodology for measuring body composition by DXA is similar to that for DPA, which has been described in detail (Peppler and Mazess, 1981; Gotfredsen *et al.*, 1984). Briefly, the measurement of body composition by this DXA system is based on the differential attenuation of low (38 keV) and high energy (70 keV) x-rays by fat and other soft tissues. The fat and lean content is determined for each pixel of a total body scan that does not overlie bone and is reported to be independent of tissue thickness. The soft tissue attenuation coefficient (R_{ST}) is the ratio of the mass attenuation coefficients (Gotfredsen *et al.*, 1986) at 38 and 70 keV:

$$R_{ST} = \mu_{ST}{}^{38}/\mu_{ST}{}^{70} = \ln(I_0{}^{38}/I^{38})/\ln(I_0{}^{70}/I^{70})$$

Calibration studies at DPX energies of 38 and 70 keV report that R_{ST} values range from 1.2 for fat to 1.4 for 100% lean.

$$\text{Lean fraction} = (R_{ST} - R_{FAT})/(R_{LEAN} - R_{FAT})$$

In addition to whole body composition values for fat and lean content, DXA measurement also estimates bone mineral content (BMC) and total mass of soft tissues (TMST).

A total of 130 male, Indian River broiler chickens were grown under common conditions from 1 to 7 d of age. At 7 d of age, the birds were randomly assigned to one of six dietary treatments (12, 21, or 30% crude protein and 0 or 1 mg triiodothyronine/kg diet) as described previously (Rosebrough and Mitchell, 1995). The birds were scanned by DXA at ages ranging from 4 to 8 wk and weights ranging from 403 to 3,289 g. For the purpose of evaluating the scan results, the birds were grouped by weight from 403 to 1,999 or 2,000 to 3,289 g. The variety of dietary treatments were employed in order to induce a wide range in body composition within each weight group. At the time of scanning the birds were either anesthetized using a combination of ketamine, xylazine, tiletamine, and zolazepam or killed by CO_2 inhalation; then placed on the instrument in a prone position and a total body scan performed. Single whole-body scans were acquired and analyzed using either pediatric total body research software (neonatal-medium mode) or small animal total body research software (detail or high resolution mode). Table 1 provides a summary of the number and weight ranges for birds scanned by the various procedures. Groups 3 and 4 consisted of a total of 54 birds. These birds were scanned live (3C) at 4, 6, and 8 wk of age, with the final scan results being included as Groups 3A and 3B. After these birds were killed, they were scanned again and reported as Groups 4A and 4B. The DXA measurements provided readings of total tissue mass, percentage fat, fat tissue mass, lean tissue mass, and bone mineral content. After scanning (except for the birds in Group 3C), the bodies of the chickens were frozen for later analysis. The frozen carcasses were defeathered, autoclaved, and then homogenized for 30 s in a food processor. Samples were freeze-dried to determine water content. The amount of protein was determined by analyzing for nitrogen by standard Kjeldahl procedures (Association of Official Analytical Chemists, 1984). Lipid content was measured by the method of Folch *et al.* (1957).

Linear regression analysis of the results and *t* test comparison of mean DXA and chemical values were performed using Statgraphics® procedures (STSC, 1992).

RESULTS AND DISCUSSION

A visual rendering of the DXA scan of a chicken is shown in Figure 1. This image illustrates the difference in contrast observed for bone and soft tissue.

[2]Lunar™ DPX-L, Lunar Corp., Madison, WI 53700.

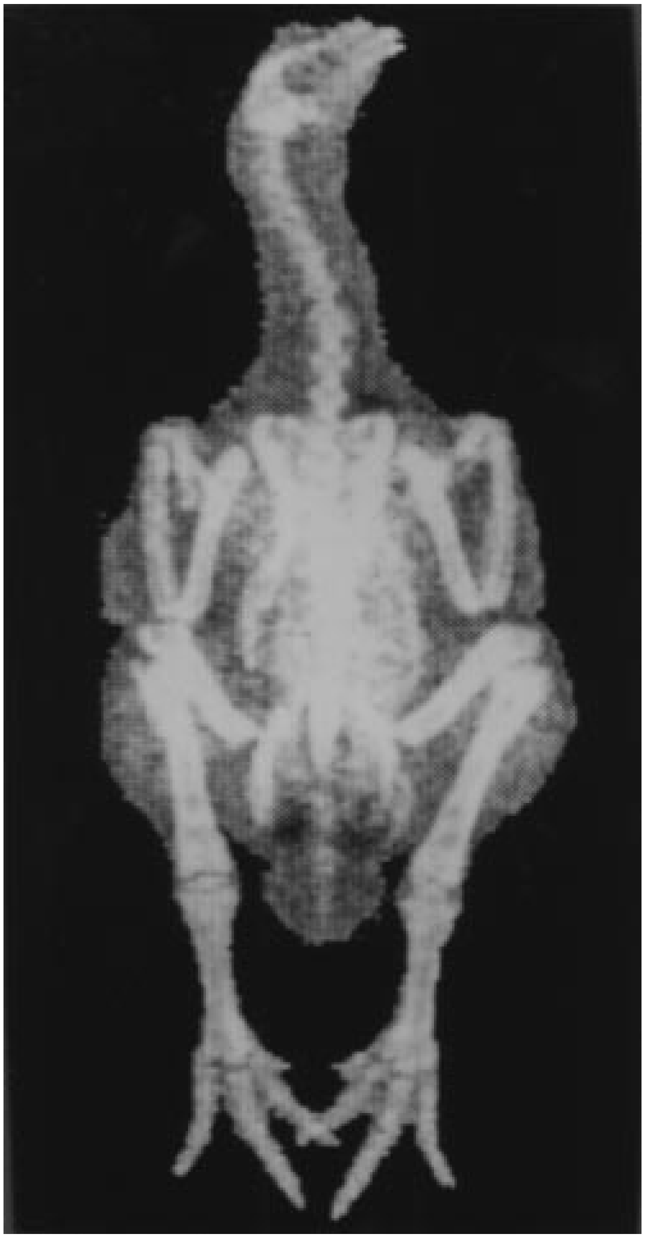

FIGURE 1. Dual energy x-ray absorptiometry scan image of a chicken.

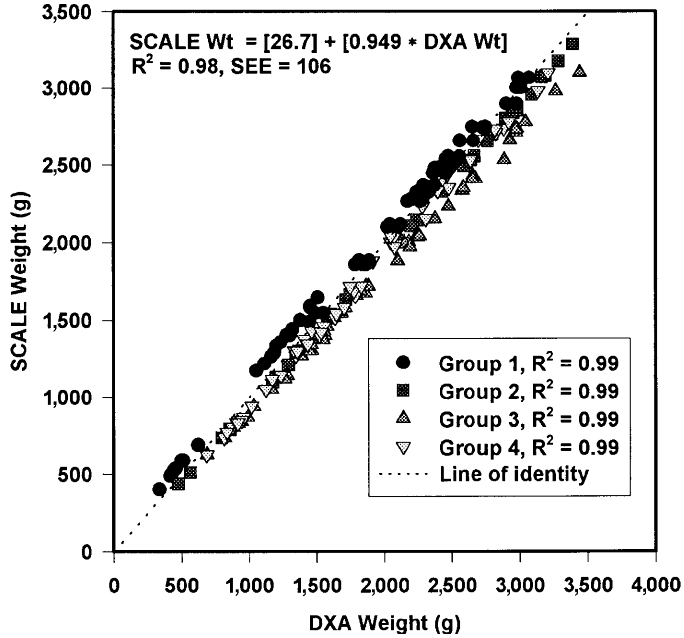

FIGURE 2. Relationship between dual energy x-ray absorptiometry (DXA) measurement of total tissue mass (DXA weight) and scale measurement of actual body weight.

Body Weight

Summation of the DXA measurements of total fat, lean body mass, and bone mineral content yield a figure that should be equivalent to total body mass. In Table 2, the mean body weight of each group of chickens at the time of scanning is compared with the mean DXA tissue mass. DXA measurements acquired and analyzed using the pediatric-neonatal program were slightly less than the scale weights (Groups 1A and 1B). However, the DXA mass measured with the small animal program, using both the detail and high resolution mode (Groups 2A–4B), were consistently greater than the scale measurements of body weight. When the difference between the scale weight and the DXA weight was expressed as a percentage of the scale weight, the greatest difference was generally in the groups of birds that weighed less than 2,000 g. Only in Group 3B was there a significant difference between the two measurements. Figure 2 shows the relationship between the DXA values for total body mass and the corresponding measurements of live body weight for all 130 chickens in this study. In all groups, the two measurements were highly correlated, with R^2 values greater than 0.98.

Body Fat

The most critical measurement for DXA as a reliable method of determining body composition is the assessment of total body fat. The relationship between the DXA R value (ratio of soft tissue attenuation coefficients) and percentage fat reported by DXA is shown in Figure 3. There was a linear relationship between the R value and estimated percentage fat for R values less than 1.385 (R^2 = 0.99). When the R value exceeded 1.385, the estimated percentage fat remained constant at approximately 4%.

The accuracy of DXA for measuring total body fat was a function of the scan program and mode and the weight of the bird. DXA and chemical measurements of total body fat are compared in Table 2. With birds weighing less than 2,000 g, DXA overestimated (P < 0.001) body fat when scanned in the pediatric-neonatal or small animal-high resolution modes (Groups 1A and 4A). However, DXA underestimated (P < 0.001) body fat in birds weighing less than 2,000 g when scanned using the small animal-detail mode (Groups 2A and 3A). The relationship between DXA and chemical measurements of percentage fat in birds

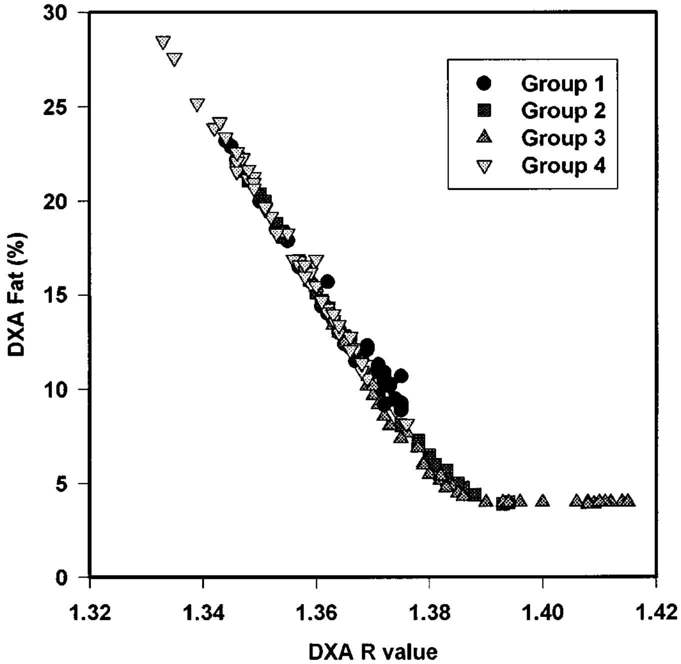

FIGURE 3. Relationship between the dual energy x-ray absorptiometry (DXA) R value and percentage body fat as measured by DXA.

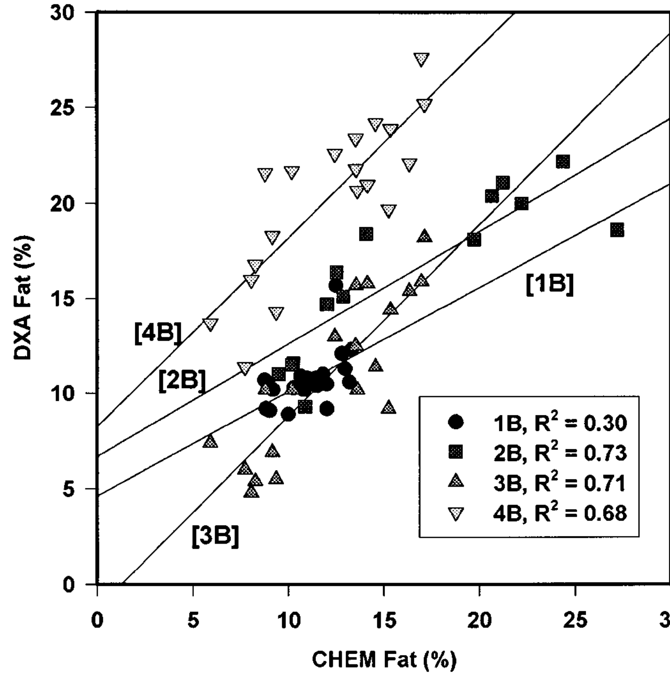

FIGURE 5. Relationship between the amount of total body fat in chickens weighing between 2,000 and 3,290 g as measured by dual energy x-ray absorptiometry (DXA) (predicted from R values) or by chemical (CHEM) analysis.

weighing less than 2,000 g is shown in Figure 4. For all groups, the correlation between percentage fat measured by the two methods was low, R^2 values ranging from 0.33 to 0.47. Mean values for DXA measurement of body fat in chickens weighing more than 2,000 g was in good agreement with chemical measurement, except for those birds scanned using the small animal-high resolution mode, which significantly ($P < 0.05$) overestimated body fat. Figure 5 shows the relationship between DXA and chemical measurements of percentage body fat of chickens weighing more than 2,000 g. With the exception of Group 1B, in which there was little variation in fat content, the correlation between the two methods was higher than with birds weighing less than 2,000 g. Combining the results of all four groups (n = 184), the correlation (R^2) between the DXA and chemical measurements for the total amount (grams) of fat in chickens was 0.62.

Although these results indicate that DXA can probably be used to provide a reliable measure of body fat in chickens; additional measurements are needed to provide a reliable prediction equation, or program algorithms may need to be modified to overcome systematic errors. Brunton *et al.* (1993), using another commercially available brand of DXA, observed that with small piglets weighing approximately 1,600 g, DXA overestimated total body fat content by more than twofold and the correlation (r) with chemically determined fat content was only 0.06. In the same study, with larger piglets (approximately 6 kg) total body fat was overestimated by only 35% and r = 0.83.

Lean Tissue

In addition to percentage and total body fat, the DXA scan provides a measure of lean body mass, which actually includes all other components of the soft tissue mass excluding fat. As with fat, the DXA lean mass value

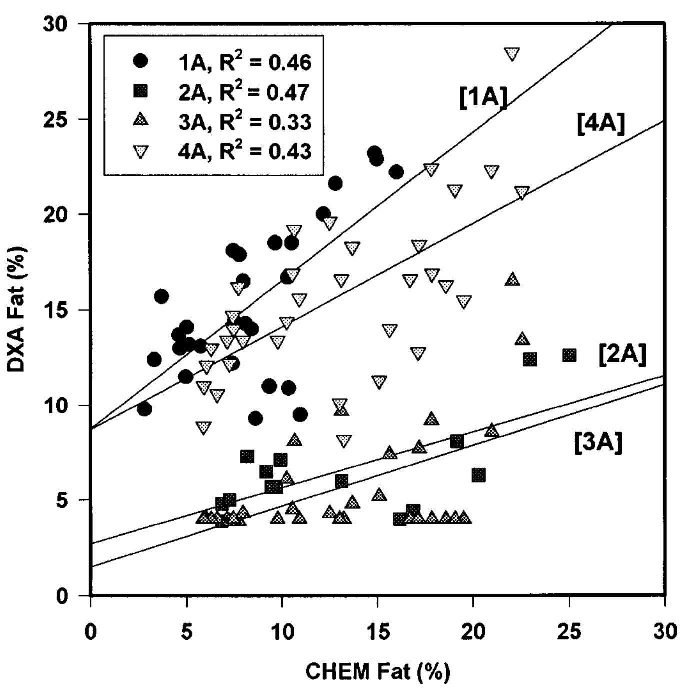

FIGURE 4. Relationship between the amount of total body fat in chickens weighing between 400 and 1,900 g as measured by dual energy x-ray absorptiometry (DXA) (predicted from R values) or by chemical (CHEM) analysis.

TABLE 2. Comparison of dual energy x-ray (DXA) measurements with chemical (CHEM) measurement of total body fat and scale measurement of total body weight of broiler chickens

Group	Body weight				Body fat				Body fat			
	Scale	DXA	R^2	SEE	Chem	DXA	R^2	SEE	Chem	DXA	R^2	SEE
	(g)				(%)				(g)			
1A	1,210 ± 89	1,126 ± 89	0.996	29	8.4 ± 0.7	15.3 ± 0.8***	0.46	2.7	95 ± 9	157 ± 9***	0.42	38
1B	2,521 ± 63	2,498 ± 70	0.984	36	11.2 ± 0.4	10.8 ± 0.4	0.30	1.3	282 ± 13	268 ± 9	0.78	27
2A	1,205 ± 108	1,377 ± 102	0.999	6	13.4 ± 1.6	6.7 ± 0.7***	0.47	4.7	148 ± 17	86 ± 11**	0.65	40
2B	2,699 ± 111	2,854 ± 116	0.999	10	16.3 ± 1.6	16.3 ± 1.2	0.73	3.3	419 ± 27	455 ± 25	0.40	81
3A	1,299 ± 64	1,426 ± 70	0.997	20	12.6 ± 0.9	5.5 ± 0.5***	0.33	4.4	149 ± 11	82 ± 10***	0.68	36
3B	2,458 ± 76	2,704 ± 83*	0.993	28	12.1 ± 0.8	11.0 ± 1.0	0.71	1.9	282 ± 21	301 ± 29	0.81	40
4A	1,299 ± 64	1,391 ± 66	0.995	18	12.6 ± 0.9	15.5 ± 0.7*	0.43	4.0	149 ± 11	219 ± 16***	0.61	40
4B	2,458 ± 76	2,533 ± 83	0.986	41	12.1 ± 0.8	20.3 ± 1.0***	0.68	2.0	282 ± 21	520 ± 35***	0.73	48

*$P < 0.05$.
**$P < 0.01$.
***$P < 0.001$.

is a function of the R value measurement. A plot (not shown) of R values vs DXA lean tissue mass expressed as a percentage of body weight revealed a positive slope inversely equivalent to the plot for percentage fat (Figure 3) with and identical R^2 value of 0.99. Body protein and water would be expected to be major components of the DXA lean body mass. Linear regression analysis revealed correlations (R^2) between the DXA R values and percentage body protein ranging from 0.02 for Group 1A to 0.57 for Group 2A with a mean of 0.28. Correlations (R^2) between the DXA R values and percentage body water were somewhat better than for percentage protein and ranged from 0.18 for Group 1A to 0.74 for Group 4B with a mean of 0.43. Thus, it does not appear that percentage body protein or water could be predicted with acceptable accuracy from DXA R values.

On the other hand, DXA lean body mass values were highly correlated ($R^2 = 0.90$ to 0.97) with total body protein as measured by chemical analysis (Table 3). The relationship between DXA lean and total body protein measurements is shown in Figure 6. Combining the data from all birds, the relationship between grams of DXA lean body mass (LBM) and grams of total body protein (P) is described by the following equation: $P = -2.2 + 0.187$ (LBM_{DXA}). Likewise, DXA lean body mass values were highly correlated ($R^2 = 0.90$ to 0.99) with total body water as measured by chemical analysis (Table 3). The relationship between DXA lean and total body water measurements is shown in Figure 7. Combining the data from all birds, the relationship between grams of DXA LBM and grams of total body water (W) is described by the following equation: $W = 33 + 0.68$ (LBM_{DXA}). Although these comparisons of DXA lean body mass and total body protein or water result in high correlations, it should be noted that the wide range in body weights had a substantial influence on these relationships. Using the predicted values for total body protein based on the above equation to calculate percentage protein, the correlation with chemically determined percentage protein was quite low, as was also the case for the correlation obtained using DXA R values to predict percentage of water.

Bone Mineral

DXA instruments are designed and marketed commercially for the clinical assessment of bone mineral status, primarily in the adult human. For that purpose, these instruments are considered to be quite accurate. In the studies reported here with chickens, the DXA scan results included measurements of bone mineral content (BMC) and density. Chemical analysis did not include bone mineral per se; however, total body ash was measured in

TABLE 3. Linear regression analysis comparing dual energy x-ray absorptiometry (DXA) lean measurements with total body protein or water and DXA bone mineral content (BMC) with total body ash

Group	DXA lean vs protein		DXA lean vs water		DXA BMC vs ash	
	R^2	SEE	R^2	SEE	R^2	SEE
1A	0.95	19.0	0.97	53.4	. . .	. . .
1B	0.93	14.0	0.95	40.5	0.05	8.9
2A	0.97	12.8	0.99	32.7	0.54	10.2
2B	0.95	20.7	0.95	82.5	0.51	19.2
3A	0.92	19.8	0.90	89.2	0.57	9.1
3B	0.91	20.6	0.95	49.3	0.23	12.5
4A	0.97	13.2	0.98	38.2	0.63	8.5
4B	0.90	22.0	0.95	49.7	0.25	12.4

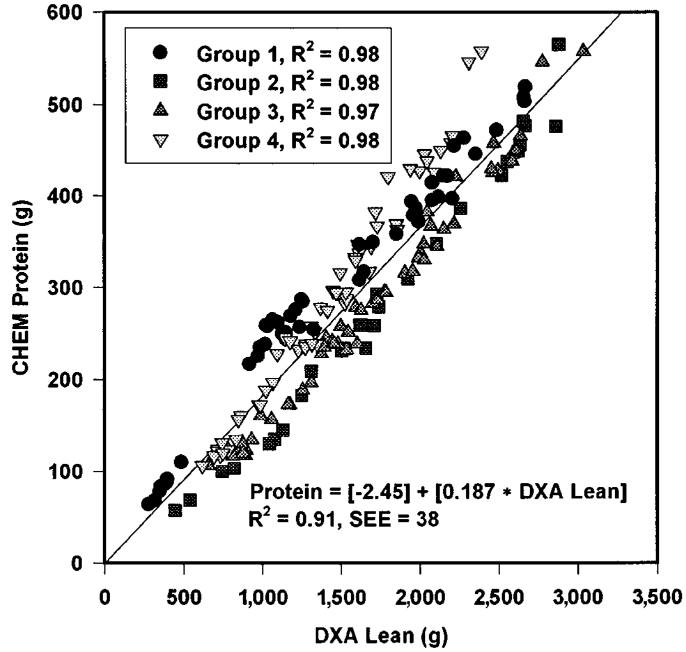

FIGURE 6. Relationship between the amount of total body protein in chickens as measured by dual energy x-ray absorptiometry (DXA) (predicted from lean body mass) or by chemical (CHEM) analysis.

all birds except those in Group 1A. Because bone mineral is the major source of total body ash, it was expected that there would be a high correlation between the DXA measurement of BMC and total body ash. However, as is shown in Table 3, this was not the case. The exact reason for the low correlation between DXA BMC and total body ash, particularly in Group 1B, is not known. When all groups were combined, the R^2 for the correlation between BMC and total body ash was only 0.46. DXA BMC and total body ash showed similar positive correlations with total body weight with R^2 values of 0.72 and 0.75, respectively. However, as shown in Figure 8, when the DXA small animal-high resolution scans (Group 4) were separated from the DXA neonatal-medium and the small animal-detail scans (Groups 1, 2, and 3) the correlations between the DXA BMC values and body weight were much higher. This figure also reveals that the high resolution mode gave higher BMC values relative to body weight than did the other two scan modes. In fact, the mean for the high resolution BMC values in Group 4 was not significantly ($P > 0.05$) different from the mean value for total ash, whereas the mean BMC values for all of the other groups were significantly less ($P < 0.05$) than the corresponding ash values. In the DXA study with small piglets (1,600 g) reported by Brunton *et al.* (1993), DXA BMC was 30% less than total body ash and the correlation (r) between the two measurements was only 0.16. The mean ratio of BMC measured by DXA and total body ash for all chickens analyzed in this study was 0.77, which is the same as observed previously (Mitchell *et al.*, 1996) for pigs analyzed at 90 kg live body weight.

Total Body Growth

The main advantage of any nondestructive method for measuring body composition is the potential for sequential assessment of changes in body composition of the live animal. DXA has been used to measure changes in the composition of pigs during growth from approximately 20 to 90 kg (Mitchell *et al.*, 1996). Figure 9 shows the composite results of the DXA measurements of live chickens (Table 1, Group 3C) during the period of 4 to 8 wk of age. These results are plotted relative to total body weight. The DXA measurements of total tissue weight (R^2 = 0.998), lean tissue weight (R^2 = 0.98), and BMC (R^2 = 0.94) increased linearly with increasing body weight. Due to the variety of dietary treatments employed, it was expected that a considerable variation in total body fat would be observed throughout the range in body weights (R^2 = 0.59). The greatest variation in body fat content was observed when body weights exceeded 2,000 g, indicating that this was where the greatest divergence in fat deposition occurred. A similar relationship between body weight and fat content was observed for the combined results of the four groups of birds (n = 184) described earlier, using either DXA (R^2 = 0.57) or chemical (R^2 = 0.55) analysis.

For the assessment of soft tissue composition, DXA instruments have been calibrated against phantoms of known fat content. Currently, the computation of soft tissue composition by these instruments is optimized for adult human subjects and can probably be improved for specific animal populations in which true body composition can be determined chemically. Furthermore, there have been no reported attempts to evaluate the lean tissue mass in terms of actual muscle mass, which would involve

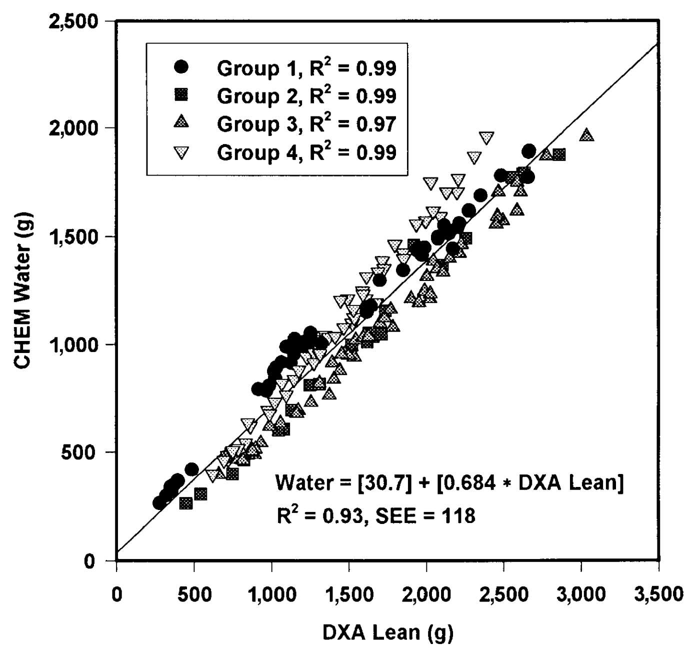

FIGURE 7. Relationship between the amount of total body water in chickens as measured by dual energy x-ray absorptiometry (DXA) (predicted from lean body mass) or by chemical (CHEM) analysis.

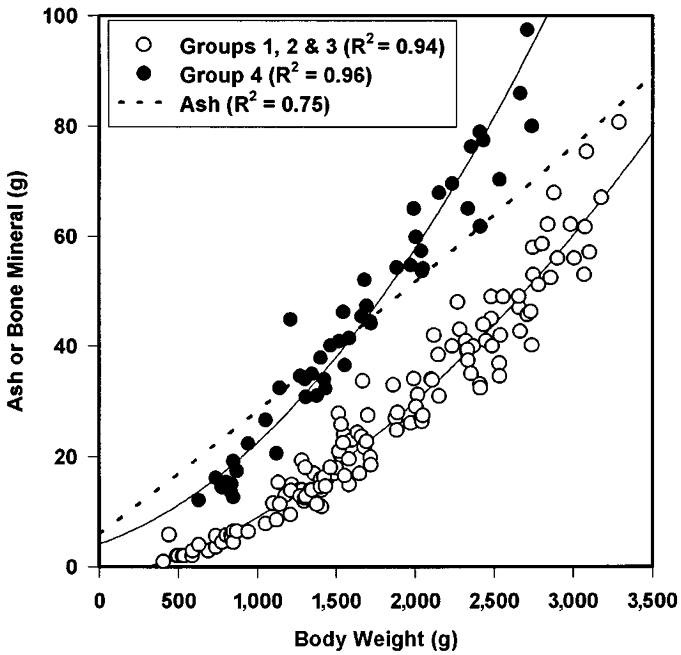

FIGURE 8. Relationship between body weights and measurements of total body ash, bone mineral by dual energy x-ray absorptiometry (DXA) neonatal-medium or small animal-detail analysis (Groups 1, 2, and 3) and bone mineral by DXA small animal-high resolution analysis (Group 4).

certain assumptions regarding the composition of skin and skeleton and the water content of adipose tissue. One of the advantages of DXA over other nondestructive methods that are being developed for measuring body composition of birds (i.e., those based on total body electrical conductivity) is the potential for measuring three components (fat, lean, and bone mineral) independent of body weight. The results of this study indicate that DXA might be appropriate for measuring the body composition of chickens weighing in excess of 2,000 g; however, there is a need for specific calibration of the DXA instrument for the measurement of body composition of chickens. It appears that for chickens weighing less that

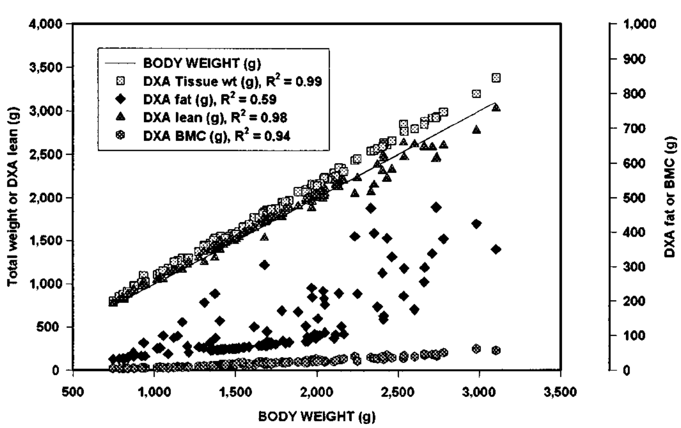

FIGURE 9. Composite results of dual energy x-ray absorptiometry (DXA) measurements of lean, fat, bone mineral (BMC), and total tissue weight in live chickens (Group 3C, Table 1) at 4, 6, and 8 wk of age.

approximately 2,000 g, there may be some fundamental problem with either the use of DXA for measuring body composition or, more likely, with the algorithms used in the calculations of body composition of small animals. Additional studies are needed to identify the nature of these problems associated with the use of DXA for measuring the body composition of chickens and to make corrections or modifications where possible. Assuming that these problems can be overcome, the remaining disadvantages for the use of DXA for measuring body composition of chickens is the time required to scan each bird and, related to that, the need to anesthetize the bird to ensure that it remains motionless during the scanning procedure.

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