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Fat mass compared to four body condition scoring systems in the Asian elephant (*Elephas maximus*)

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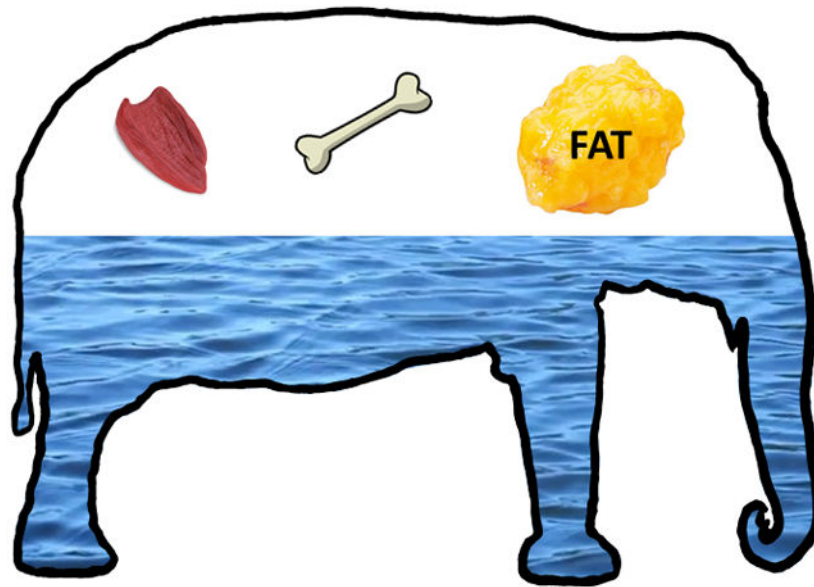
Abstract

Captive elephant populations are not self-sustaining due to health concerns possibly related to obesity. Categorizing obesity relies on qualitative analyses like body condition scores (BCS). However, elephant indices have not been validated against measured body composition. The objective was to compare BCS systems to body composition determined by deuterium dilution in 28 zoo-kept Asian elephants. Elephants were weighed and given deuterated water orally (0.05 mL/kg). Blood was collected at ~0, 24, 120, 240, 360, and 480 h after dosing. Photographs were taken to score the elephant based on four BCS systems (BCS_{Wemmer} [0 to 11 scoring], BCS_{Morfeld} [1 to 5 scoring], BCS_{Fernando} [0 to 10 scoring], BCS_{Wijeyamohan} [1 to 10 scoring]). Based on regression analysis, relative fat ranged from -305 kg to 515 kg, where negative values indicate less and positive values indicate more fat than expected for the elephant's mass in this population. BCS_{Fernando} was associated with relative fat ($P = 0.020$, $R^2 = 0.194$). Relative fat, adjusted for sex and age in the statistical model, was associated with BCS_{Wemmer} ($P = 0.027$, $R^2 = 0.389$), BCS_{Fernando} ($P = 0.002$, $R^2 = 0.502$), and BCS_{Wijeyamohan} ($P = 0.011$, $R^2 = 0.426$). Inclusion of zoo and familial relatedness resulted in all BCS systems associated with relative fat ($P < 0.015$). Only BCS_{Fernando} predicted relative fat, unadjusted, suggesting it is the most capable system for practical use. Compared to absolute fat, relative fat may be more biologically relevant as greater fat relative to body mass is more likely to lead to health issues.

Graphical Abstract

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Keywords

body composition; BCS; zoo

Introduction

Free-ranging elephant populations are rapidly declining, so captive breeding can be one means of insurance against extinction (Conde, Flesness, Colchero, Jones, & Scheuerlein, 2011; Hoffmann et al., 2010). However, most captive populations are not self-sustaining, and over the past decade, mortality rates have been greater than birth rates for elephants housed in North American zoos (Faust & Marti, 2011). This problem is in part due to logistics, as males are often not housed with reproductive females (Holt, Brown, & Comizzoli, 2014), but also to a high prevalence of reproductive and health issues, including arthritis, dystocia, and abnormal ovarian cycles (Brown, 2000; Clubb & Mason, 2002; Lewis, Shepherdson, Owens, & Keele, 2010), some of which may be related to excess fat.

In a recent survey of elephants housed in American Zoo and Aquarium (AZA) accredited zoos, 75% of female and 65% of male Asian elephants were characterized as being overweight or obese (Morfeld, Meehan, Hogan, & Brown, 2016). Similar findings have been shown in Asian elephants housed in European institutions (Schiffmann et al., 2018).

However, assessing body condition is generally based on a qualitative body condition score (BCS), rather than quantified measures of total body fat. There are several BCS systems for Asian elephants, each involving a qualitative evaluation of key skeletal descriptors (e.g., ribs, pelvic bone, backbone) either by direct observation or via photographs (Morfeld et al., 2016; Wemmer et al., 2006). Based on the appearance of these anatomical regions, the elephant is given a numerical score, with lower numbers representing less and higher numbers representing more subcutaneous fat (Morfeld et al., 2016). Higher BCSs have been shown to be associated with increased ovarian acyclicity and changes in lipid, metabolic and adrenal

status (Morfeld & Brown, 2014; Norkaew et al., 2018), as well as being a non-breeding female elephant, irrespective of species (Schiffmann et al., 2018). Although BCSs correlate positively with measures of subcutaneous fat (Morfeld, Lehnhardt, Alligood, Bolling, & Brown, 2014; Treiber, Reppert, & Ward, 2012), it has not been determined that BCSs accurately reflect the body composition of elephants.

The BCS technique was originally developed to assess the soft tissue (i.e., muscle and subcutaneous fat) of livestock to evaluate their nutritional and economic efficiency (Jefferies, 1961). However, BCSs are often extrapolated to infer a level of fatness, rather than soft tissue, which deviates from its original purpose with little quantitative supportive data. To date, elephant BCS protocols have been compared to serum triglycerides (Morfeld et al., 2016) and ultrasound measures (Morfeld et al., 2014), but neither are a measure of body composition. Although Morfeld (2016) found a positive relationship between BCSs and serum triglycerides, it is widely established that serum triglycerides are strongly influenced by diet (Liu et al., 2015; Mensink & Katan, 1992). In addition, triglycerides are not produced by adipocytes (fat cells). Ultrasound units are capable of estimating subcutaneous fat mass thickness (measured in millimeters not grams or kilograms), but in the elephant, they cannot account for visceral and ectopic fat, thereby not providing a measure of body composition. Further, the reliability of BCSs is contingent on the variation among and within the individuals scoring the elephant (Dugdale, Curtis, Milne, Harris, & Argo, 2011; Schiffmann, Clauss, Hoby, & Hatt, 2017). Therefore, prior to accepting BCSs as a valid means of estimating fatness in elephants, it is helpful to compare BCSs to a more direct measure of total body fat (Charette, Bigras-Poulin, & Martineau, 1996).

The best means to estimate total body fat mass (FM) in large species, such as the elephant, is by deuterium dilution. Deuterium dilution is a non-destructive technique that measures the animal's total body water, which is then used to estimate fat free mass (FFM) (Wang, Pierson, & Heymsfield, 1992). Fat mass is calculated as the difference between body mass and FFM. Deuterium dilution has been validated in a range of animals, from bumblebees to the Atlantic walrus (Acquarone & Born, 2007; Wolf, Ellington, Davis, & Feltham, 1996), and we have previously shown the feasibility of using this method to measure body fat in African elephants (Chusyd et al., 2018). The primary objective of this study was to compare four commonly used BCS systems with FM estimated by deuterium dilution to determine which scoring system most accurately reflects FM in both female and male zoo Asian elephants.

Methods

Animals

This study was approved by the Institution Animal Care and Use Committee of the University of Alabama at Birmingham (UAB), the Smithsonian Conservation Biology Institute (SCBI), and participating zoos. A total of 28 elephants participated in the study (females: n=23 from seven zoos, mean age 31 ± 3.0 years, age range 8 – 56 years; males: n=5 from five zoos; mean age 21 ± 5.4 years; age range 8 – 34 years). Female elephants were not pregnant, but four had calves that ranged from 1.5 to 4.5 years of age. Male elephants were not in musth at the time of the study.

Body Composition

Body composition was assessed as previously described (Chusyd et al., 2018). In brief, using the institutions' scales, elephants were weighed to the nearest 1 or 5 pounds. Zoo personnel collected venous blood from an ear or leg vein (~9 mL) to determine background isotope enrichment before administering deuterated water. The location of blood collection remained the same for subsequent samples by elephant. An oral dose of deuterium oxide (0.05 mL D₂O/kg of body mass; 99.9% APE; DLM-4-1000, Cambridge Isotopes, Tewksbury, MA) was administered using bread (Publix®, Birmingham, AL) as the vehicle. Post deuterium administration, blood (~9 mL) was collected at regular intervals (~24, 120, 240, 360, and 480 h). Whole blood was centrifuged within 30 minutes to separate serum. Serum was aliquoted, and frozen at a minimum of -20 °C until shipped on dry ice overnight to UAB. Samples were kept in airtight containers in a frost-free -80 °C freezer until analysis.

Isotope ratio mass spectroscopy (Finningan Delta V Advantage, Thermo Fisher Scientific, USA) analysis was carried out by UAB's Nutrition Obesity Research Center's Metabolism Core with guidance and support from the Energetics Research Group at the University of Aberdeen, Aberdeen, Scotland (Chusyd et al., 2018). In brief, the 2H/1H delta value was converted to parts per million, and used to calculate FFM based on the mammalian hydration constant (0.73) (Speakman, 1997). Fat free mass was then subtracted from body mass to infer FM.

Body Condition Score (BCS)

The BCS system developed by Morfeld and colleagues (Morfeld et al., 2016), BCS_{Morfeld}, is based on three anatomical regions (ribs, pelvic bone, and backbone) from various angles (e.g., lateral, rear-angle, and rear view) using a 1- to 5-point scoring system by whole numbers only. The BCS system developed by Wemmer and colleagues (Wemmer et al., 2006), BCS_{Wemmer}, is based on six anatomical regions (head, scapula, thoracic region, the area in front of the pelvic bone, backbone, pelvic bone) from various angles (e.g., lateral, rear, and elevated views) using a 0- to 11-point scoring system by 0.5 increments. The BCS system developed by Fernando and colleagues (Fernando, Janaka, Ekanayaka, Nishantha, & Pastorini, 2009), BCS_{Fernando}, compares the test elephant to five reference photographs preassigned a score of 1, 3, 5, 7, and 9 from a lateral view only. The test elephant is given one of the preassigned scores if it looks like the elephant in the reference photograph, or if it falls between the reference photographs, an even score is given, resulting in a scoring system from 0 to 10 by whole numbers only. The BCS system developed by Wijeyamohan and colleagues (Wijeyamohan, Treiber, Schmitt, & Santiapillai, 2015), BCS_{Wijeyamohan}, is based on reference photographs coupled with an explanatory key, scoring elephants from 1 to 10 by whole numbers only based on the lateral view of the elephant. For each index, lower scores imply less fat and higher scores imply more fat.

For each elephant, a set of photographs was taken by an observer around the elephant from every 45° angle along the horizontal plane (8 photographs per elephant beginning with a frontal view of the elephant), on the same day deuterated water was administered, to score body condition using each of the four BCS indices. To assess intra- and inter-assessor

variability, three assessors scored each elephant three times, with a minimum of 1 week between scoring. Photographs were randomized prior to each scoring session. Scores were generated by the author (DEC) and two assessors who were trained by DEC. Intraclass correlations (ICC (2,1)) were done to evaluate intra- and inter-assessor variability. The first round of scoring showed the strongest intra-assessor reliability, $ICC(2,1) = 0.655 - 0.831$. Therefore, BCSs from the first round of scoring were averaged across assessors to determine the final BCSs for each elephant. There were no significant effects on the primary model outcomes when BCSs were used exclusively from one assessor's scoring or from other rounds of scoring.

Statistical analyses

Statistical analyses for the primary models were performed using SAS v9.4 statistical software (SAS Institute, Cary, NC, USA), while secondary sensitivity analyses were performed using R (R Development Core Team, 2008). All statistical analyses were determined prior to examining the data unless otherwise stated. Although body composition was conducted on 30 elephants, two elephants (1 male, and 1 female) were excluded because they did not ingest their total deuterated water amount; thus, it was not possible to determine the exact amount ingested. Therefore, calculating body composition accurately was not possible. Therefore, all models included 28 elephants.

The primary models to address the main hypothesis were linear models regressing relative fat on each BCS system, with subsequent analyses including FM, FFM, and body mass. The BCS systems differed in the number of scores that can be given, thus it was predicted those systems with a more differentiated range would have higher R^2 values in regard to quantitative measures. Sex and age were included in the primary model as covariates, followed by secondary sensitivity analyses including zoo. Linear mixed models regressed relative fat, FM, FFM, or body mass on each BCS system, with familial relationships treated as random effects. To address familial relationship, an R package called *pedigreemm* was used that allows for the correlations of genetic relationships by taking into account the variation within and between sire and dam (Vazquez, Bates, Rosa, Gianola, & Weigel, 2010). Out of 15 related pairs, 1 was a full sibling pair, 5 were half sibling pairs, and 9 were parent offspring pairs. In addition, 9 out of the 15 pairs resided at the same zoo. All model results included an Akaike Information Criterion (AIC) score to determine which model was the best fit based on the lowest score.

Relative FM was determined by the residual for each elephant when FM was regressed on body mass as done in other publications (Franco-Villoria et al., 2016; Goran et al., 1998; Secor & Nagy, 2003). Relative FM (i.e. the residual) is the amount of fat above or below their expected value after taking into account body size (i.e., body mass). As total FM and FFM typically increase with body size, this outcome variable is likely more biologically relevant than absolute FM. Shapiro-Wilk test was used to test normality for the residuals and was not significant ($P=0.301$).

Descriptive statistics were assessed for the total data set, and then by sex. Body fat percent was included as a descriptive statistic, but not as an outcome variable. Body fat percent is a ratio, and as such, may not be appropriate as an outcome variable in hypothesis testing.

Specifically, to control for the denominator (i.e., body mass), the intercept of the regression of the numerator (i.e., fat mass) on the denominator needs to be zero (Allison, Paultre, Goran, Poehlman, & Heymsfield, 1995). Our data did not satisfy this requirement. Nonparametric testing was used when comparing by sex as the body composition data for males were not normally distributed. Significance was set at $P < 0.05$ (2-tailed).

Results

Descriptive statistics for the entire population and by sex are presented in Table 1. BCSs for each elephant by BCS system are presented in Table 2.

The primary models investigated the relationship between each BCS system and relative fat, with subsequent models investing absolute FM, FFM, and body mass (Table 3). Only BCS_{Fernando} was significantly associated with relative FM (Figure 1). BCS_{Wemmer} , BCS_{Fernando} , and $BCS_{\text{Wijeyamohan}}$ significantly predicted absolute FM (Figure 2). BCS_{Morfeld} was not significantly associated with either outcome.

The primary models were then adjusted for sex and age (Table 4). BCS_{Wemmer} , BCS_{Fernando} , and $BCS_{\text{Wijeyamohan}}$ were significantly associated with relative FM. All BCS systems were significantly associated with absolute FM, and body mass. BCS_{Wemmer} , BCS_{Fernando} , and $BCS_{\text{Wijeyamohan}}$ were significantly associated with FFM, while BCS_{Morfeld} almost reached significance. The models were also adjusted for sex only (Table S1) and age only (Table S2) to determine possible effects of just one covariate versus the other.

Results after accounting for zoo and familial relatedness in the model with age and sex are presented (Table 5). All BCS systems were associated with relative FM and absolute FM measures. Only BCS_{Wemmer} was significantly associated with FFM. BCS_{Wemmer} , $BCS_{\text{Wijeyamohan}}$ and BCS_{Fernando} were significantly associated with body mass. The models were also adjusted for zoo only (Table S3) and familial relatedness only (Table S4) to determine possible effects of just one covariate versus the other.

AIC score was included to determine the best fitting models on each outcome. For all BCS systems, and all outcomes, the best model adjusted for sex, age, zoo, and familial relatedness. For relative FM, BCS_{Wemmer} , BCS_{Fernando} , and $BCS_{\text{Wijeyamohan}}$ models resulted in similar AIC scores, which were lower (i.e., better fit) than BCS_{Morfeld} .

Intra-assessor reliability ranged for BCS_{Morfeld} , BCS_{Wemmer} , BCS_{Fernando} , and $BCS_{\text{Wijeyamohan}}$, $ICC(2,1) = 0.76-0.91$, $0.83-0.97$, $0.76-0.97$, and $0.78-0.97$, respectively. Inter-assessor reliability for BCS_{Morfeld} , BCS_{Wemmer} , BCS_{Fernando} , and $BCS_{\text{Wijeyamohan}}$, $ICC(2,1) = 0.58-0.74$, $0.77-0.83$, $0.60-0.82$, and $0.59-0.66$, respectively.

Discussion

To our knowledge, this is the first study to examine how different BCS systems correspond to measures of adiposity in elephants. BCS_{Fernando} was the most reliable system to predict relative FM, unadjusted. Relative fat refers to the amount of fat an individual has after body mass differences are accounted for, as larger individuals typically have more fat overall. In

contrast, absolute fat refers to the total amount of fat the elephant has regardless of their size. Compared to absolute fat, relative fat may be more biologically relevant as greater fat mass relative to body mass is more likely to be linked to health issues associated with excess fat.

The use of deuterium dilution to quantify body composition/adiposity is a major strength of this study. Deuterium, a non-radioactive isotope of hydrogen, replaces hydrogen in water molecules, allowing the measurement of total body water (Pace & Rathbun, 1945). There is a relationship between total body water and FFM in mammals, termed the hydration constant, ultimately allowing for body composition quantification (Wang et al., 1999). Although assumptions were made (e.g., appropriate hydration constant used) and deuterium dilution has not been validated by total carcass analysis in Asian elephants, the method appears to be robust over time and species (Acquarone & Born, 2007; Burkholder & Thatcher, 1998; Cowan, Robinson, Greenhalgh, & McHattie, 1979; Dugdale et al., 2011; Farley & Robbins, 1994; Schloerb, Friis-Hansen, Edelman, Solomon, & Moore, 1950).

Relative FM is arguably the most important biological outcome when using a BCS system. Therefore, the primary linear regression model tested whether each BCS system could predict relative FM of the elephant. The Fernando system was the only system able to independently predict relative FM. A potential explanation for the Fernando system's success may be related to the view of the elephant used for scoring. The Fernando scoring system, unlike the Morfeld and Wemmer systems, only relies on the lateral view of the elephant. Although the Wijeyamohan system relies only on the lateral view, differences in results may be due to the reference animals used and the simplicity of the Fernando system. The Fernando system uses five photographs aligned in one vertical column, progressing in body condition. The reference photographs used may have had greater consistency in subcutaneous fat changes with the increasing predefined BCS and allowed for the entire spectrum of scores to be visualized. Further, the lateral view may encompass the specific fat deposits that primarily expand during positive energy balance. To determine whether this was the case, we conducted a stepwise regression analysis to predict relative FM. The stepwise regression included six anatomical regions provided by the Wemmer system, in addition to the surface area of the elephant's side region between the front and back legs. The six anatomical regions provided by the Wemmer system included the three anatomical regions used by the Morfeld system. Of the six anatomical regions used (temporal depression in the head, pronouncement of the scapula, visibility of the ribs, depression in front of the pelvic bone, visibility of the lumbar vertebrae viewed from behind the elephant, and visibility of the pelvic bone) and the surface area, the stepwise selection resulted in a model with only one explanatory variable, the ribs (AIC = 284.58). Subcutaneous fat over the ribs has not been measured via ultrasound due to practical limitations in locating the ribs reliably (Morfeld et al. 2014). The other anatomical regions relied upon may reflect anatomical changes associated with age rather than nutritional status as inferred by the wet/dry season. For example, Albl (1971) took a series of direct body measurements to investigate their relationship with subcutaneous fat and muscle mass in wild African elephants. Albl (1971) found that most of the direct measurements were indicative of age and not nutritional status as inferred based on the dry versus wet season. Of the relevant measurements, the temporal dent and the scapular depression showed a greater association with age. Further, in other species, older age is associated with increased muscle loss

(Colman, McKiernan, Aiken, & Weindruch, 2005; Deschenes, 2004), particularly in females (Janssen, Heymsfield, Wang, & Ross, 2000). The clear pronouncement of certain anatomical regions may be related to preferential muscle loss. Collectively, these results suggest that in theory, scoring only the ribs may be required when interested in elephants' relative fat stores.

When age and sex were accounted for in the primary model, the Fernando, Wemmer, and Wijeyamohan systems significantly predicted relative FM. Following exploratory analyses, it appeared that the relationship between relative FM and these systems was mediated through sex. Elephants exhibit sexual dimorphism, with males being much larger and heavier compared to their female counterparts, yet BCS systems have been generalized to either sex. The inclusion of both males and females in this study provided the opportunity to demonstrate that there may be inherent sex differences within BCS systems, albeit relying on a small sample size of males. The four included BCS systems assumed the criteria used to score the elephant are the same for males and females; however, this may not be appropriate, particularly when the scorer lacks extensive experience and may not be able to recognize subtle differences in developed musculature from fat deposits. Similar to other species (Wells, 2007), in this study sample, males overall have significantly greater FFM and relatively less FM compared to females. In addition, the majority of males tended to have less FM than expected (i.e., relative FM values for males fell below the regression line). This is because, in most mammalian species, relative to females, males have proportionately less fat (Ledger & Smith, 1964; Pitts & Bullard, 1968; Schoenemann, 2004; Wells, 2007). Therefore, it is feasible that a male could be scored a BCS =4 due to their greater FFM deposition obscuring bone structures, while a female could be scored a BCS =4 due to their greater FM obscuring bone structures. Both elephants receive the same score, but have overall different body compositions, which was recently independently posited by other investigators (Schiffmann et al., 2018).

The study population resided at eight different zoos and included 15 pairs of related individuals, the majority being a parent offspring pair (2 father offspring, 7 mother offspring), followed by half-sibling pairs. Therefore, zoo and family relatedness were included in the model to account for potentially correlated residuals attributed to the environment and genetic relatedness. BCS_{Wemmer} , $BCS_{Fernando}$, and $BCS_{Wijeyamohan}$ models resulted in similar AIC scores, which were lower (i.e., better fit) than $BCS_{Morfeld}$ when predicting relative FM (Table 5). Therefore, visual BCS may be an appropriate tool for physical monitoring of zoo elephants when focusing on elephant health. Following exploratory analyses, it was determined that zoo accounted for much of the variability and helped isolate the effect of BCS on relative FM. Therefore, within a zoo, there may be a greater correlation between BCS and relative FM. Although from a statistical and research perspective it is important to know an elephant's age, sex, familial relatedness, and housing institution when assigning a BCS, from a practical standpoint, a zookeeper cannot adjust for such factors when scoring their own elephants, thus they should focus on consistency when using a BCS system.

The Wemmer system consistently produced the highest ICC values, both in terms of intra- and inter-assessor reliability, while the Wijeyamohan system typically had the lowest. The

stronger correlation between the Wemmer scores is likely attributed to the detailed and clear description of the anatomical regions, each of which is scored separately and then the scores added, which is supported by a similar composite BCS system used in black rhinoceros (Reuter & Adcock, 1998). In comparison, the Wijeyamohan method directly compares the focal elephant photograph to a series of reference photographs, with an accompanying description. However, the reference photographs for the Wijeyamohan system are placed on multiple pages throughout the publication, making comparisons more difficult. Our results contrast with recent findings from Schiffmann et al. (2017), which found the Wemmer system to have the highest inter-assessor variability, and Wijeyamohan to have some of the lowest inter-assessor variability. Differences in results may be attributed to Schiffmann et al. (2017) modifying the scoring systems, or to the differences in the background of the scorers. For example, Schiffmann et al. (2017) treated the Wijeyamohan system as a flow chart algorithm approach, providing more detail and direction in assigning scores. This suggests that the improved consistency in scoring is likely attributed to following the flow chart for this specific system, as in the present study, the Wijeyamohan system was used based on comparisons to example photographs only. Ultimately, descriptors must be clearly defined to allow assessors certainty of their interpretations (Teasdale & Jennett, 1974), and this ambiguity may have led to the lower ICC results. Scoring accuracy may also be contingent upon the pictures each BCS system uses as their examples. Although all four systems relied on example photographs in some capacity, the lighting, background, and amount of the photograph taken up by the elephant all varied. Lack of uniformity between and within each system may have contributed to how each elephant is scored. Ultimately, the homogeneity of the photographs may prove easier for the assessor to consistently score the elephant and future systems should consider the standardization of photographs. Lastly, a flexible scoring range seemed beneficial for the systems. For example, the Wemmer, Fernando, and Wijeyamohan BCS systems score the elephants on a minimum 10-point scale, while the Morfeld system only scores elephants on a 5-point scale. By having a smaller range of scores, the Morfeld system provides less differentiation and flexibility in assigning elephants to each score. Indeed, when examining the distribution of scores in this study, there was substantial overlap between elephants scored a 4 and those scored a 5, based on the inter-assessor data. Future inclusion of half scores in the Morfeld system should improve the differentiation between elephants.

In conclusion, this study suggests that BCS better explains relative FM than absolute FM, FFM, or body mass, with the Fernando system proving to be the most reliable system to use and a wider scoring range improving the overall predictability of body fat. The success of the Fernando system may be related to only using a photographic guide based on the lateral view of the elephant showing a clear progression in the loss of visibility of the ribs. In addition, by using multiple assessors to score each elephant multiple times, it was possible to examine which BCS system proved most consistent. The Wemmer system produces the most consistent scores. This is valuable information as BCS is a tool widely used by individuals of various backgrounds. Because only elephants under human care were used in this study, it is not known whether the results carry over to free-ranging populations. Nevertheless, this was the first step required in determining the validity of BCS systems for elephants. Further, when considering health implications and/or comparing individuals that

vary in size, it is better to predict relative fat rather than absolute fat. Future development or refinement of current BCS systems should include only those measures (i.e., the ribs) that were significantly associated with relative FM as this could allow for clearer fat classifications. It is helpful to have a valid BCS system as BCSs are consistently used in Asian elephant husbandry, welfare, and research. Consistently using the same BCS system will allow keepers to recognize changes in elephants that may require intervention to improve overall wellness.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

Acknowledgments

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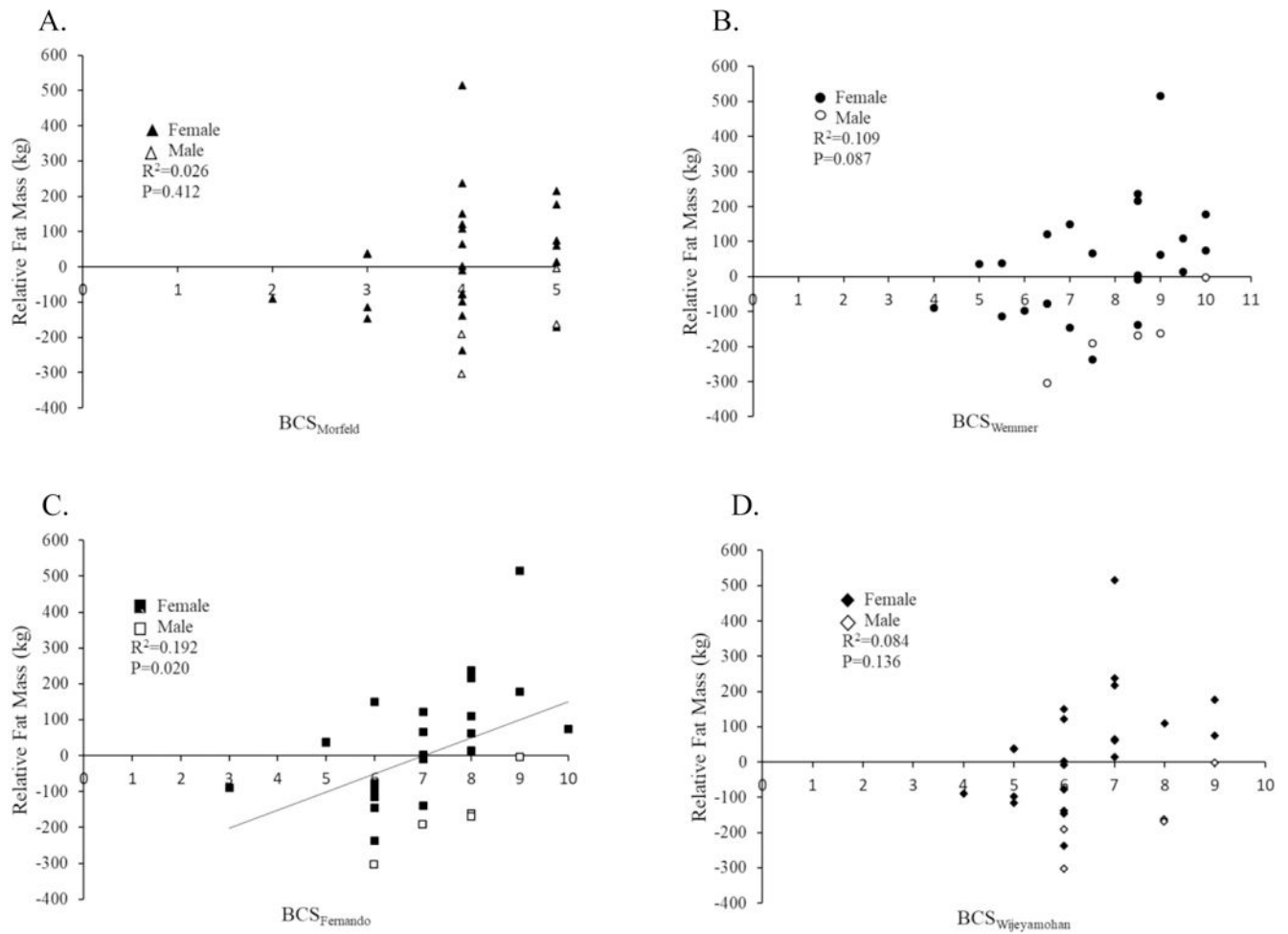


Figure 1. Distribution of Relative Fat Mass by the Morfeld BCS system (A), Wemmer BCS system (B), Fernando BCS system (C), and Wijeyamohan BCS system (D). Positive relative FM values imply the elephant has more fat than expected for their body mass, while negative relative FM values imply the elephant has less fat than expected for their body mass. Trendline indicates statistical significance.

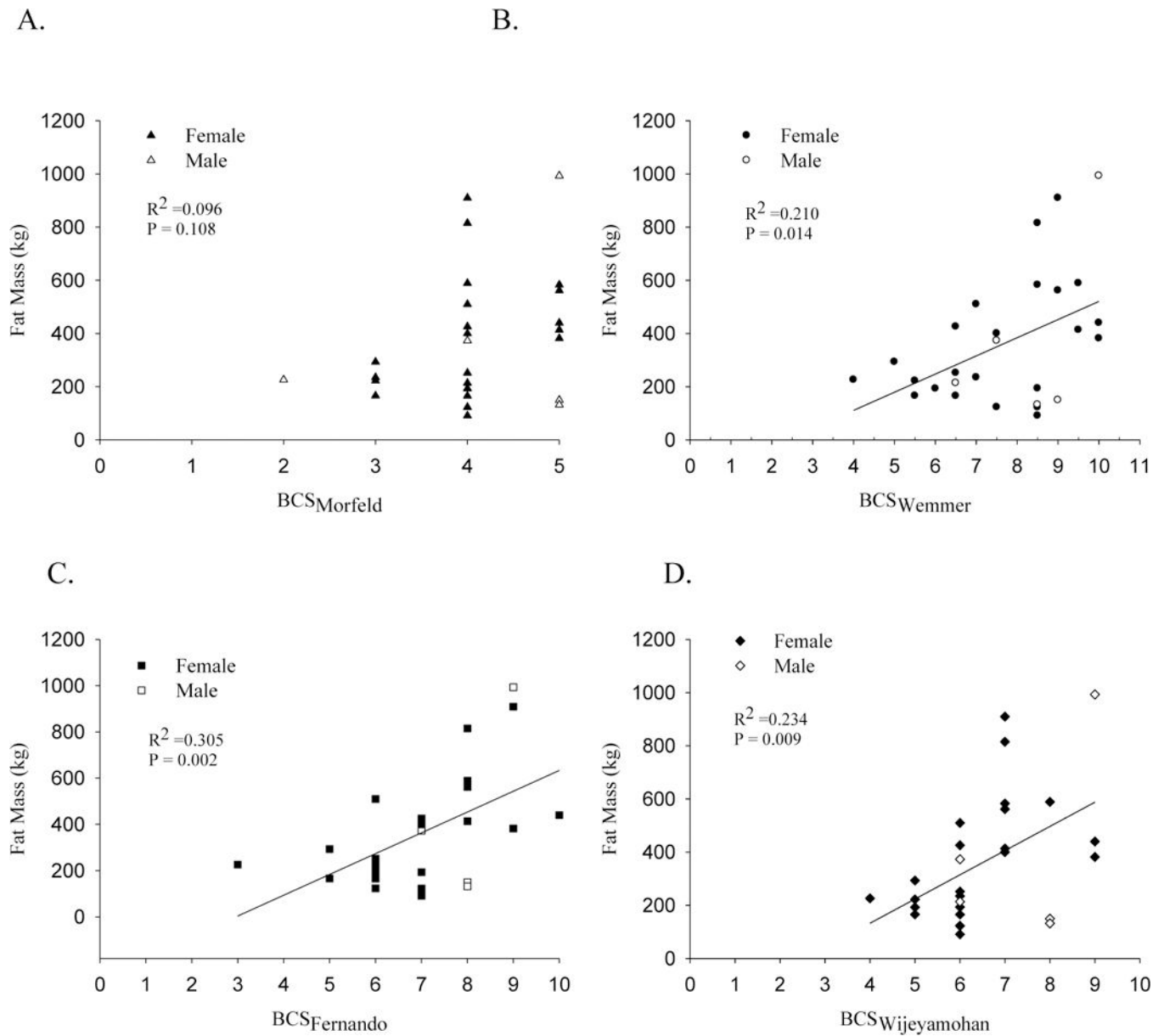


Figure 2. Distribution of absolute FM, unadjusted, by the Morfeld BCS system (A), Wemmer BCS system (B), Fernando BCS system (C), and Wijeyamohan BCS system (D). Trendline indicates statistical significance.

Table 1.Sample characteristics of the study sample (Mean \pm Standard Deviation).

	Total Sample N=28	Females N=23	Males N=5
Age (years)	29.14 \pm 14.42	31.00 \pm 14.44 ^a	20.60 \pm 12.03 ^b
Mass (kg)	3506.25 \pm 1055.86	3272.74 \pm 712.45 ^a	4580.40 \pm 1725.61 ^b
Fat Free Mass (kg)	3142.89 \pm 899.71	2911.35 \pm 578.11 ^a	4208.00 \pm 1378.19 ^b
Fat Mass (kg)	363.39 \pm 242.95	361.43 \pm 221.12	372.40 \pm 359.68
Body Fat (%)	9.91 \pm 5.01	10.54 \pm 5.07	7.01 \pm 3.88
BCS _{Morfeld} (1–5)	4.07 \pm 0.77	3.96 \pm 0.77 ^a	4.60 \pm 0.55 ^b
BCS _{Wemmer} (0–11)	7.70 \pm 1.63	7.57 \pm 1.68 ^a	8.30 \pm 1.35 ^b
BCS _{Fernando} (0–10)	7.00 \pm 1.49	6.87 \pm 1.55 ^a	7.60 \pm 1.14 ^b
BCS _{Wijeyamohan} (1–10)	6.54 \pm 1.29	6.35 \pm 1.23 ^a	7.40 \pm 1.34 ^b

Different letters represent significant differences within the row. P < 0.05.

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Table 2.

Body composition by deuterium dilution and BCSs for each elephant.

ID	Sex	Age (years)	Mass (kg)	FFM (kg)	FM (kg)	BF%	BCS _M (1–5)	BCS _W (0–11)	BCS _F (1–10)	BCS _{Wi} (1–10)
201	F	50	3343	3121	222	6.65	3	5.5	6	5
202	F	19	3699	2789	910	24.59	4	9.0	9	7
203	F	21	3483	2974	509	14.64	4	7.0	6	6
204	F	45	4345	3783	562	12.93	5	9.0	8	7
205	F	45	3611	3376	235	6.51	3	7.0	6	6
206	F	41	4819	4004	815	16.91	4	8.5	8	7
207	F	56	2854	2561	293	10.27	3	5.0	5	5
208	F	22	3313	3120	193	5.84	4	8.5	7	6
209	F	34	3733	3319	414	11.08	5	9.5	8	7
210	F	24	2089	1966	123	5.87	4	8.5	7	6
211	M	8	3198	3048	150	4.68	5	9.0	8	8
212	F	15	3520	3080	440	12.51	5	10.0	10	9
213	F	8	2538	2156	382	15.05	5	10.0	9	9
214	F	42	4216	3627	589	13.97	4	9.5	8	8
215	M	8	3128	2996	132	4.23	5	8.5	8	8
216	M	29	7382	6389	993	13.45	5	10.0	9	9
217	F	29	3484	3361	123	3.54	4	7.5	6	6
218	F	18	2762	2597	165	5.99	4	6.5	6	6
219	F	28	3526	2943	583	16.53	5	8.5	8	7
220	M	24	4740	4367	373	7.88	4	7.5	7	6
221	F	11	2064	1898	166	8.04	3	5.5	5	5
222	F	10	1823	1732	91	4.99	4	8.5	7	6
223	F	46	3062	2869	193	6.30	4	6.0	6	5
224	F	46	3329	2929	400	12.02	4	7.5	7	7
225	F	21	3300	3048	252	7.63	4	6.5	6	6
226	F	36	3146	2720	426	13.54	4	6.5	7	6
227	F	46	3214	2988	226	7.04	2	4.0	3	4
228	M	34	4454	4240	214	4.80	4	6.5	6	6

Sex: F=female, M=male; FFM: Fat free mass; FM: Fat mass; BF%: Body fat percent; BCS_M: BCS_{Morfeld}; BCS_W: BCS_{Wemmer}; BCS_F: BCS_{Fernando}; BCS_{Wi}: BCS_{Wijeyamohan}.

Table 3.

Estimates for each BCS system in statistical models to predict body composition.

Model	AIC	R ²	Estimate	SE	P
Rel. FM = BCS_M	371.64	0.03	36.03	43.19	0.412
Rel. FM = BCS_W	369.16	0.11	34.63	19.43	0.087
Rel. FM = BCS_F	366.41	0.19	50.29	20.22	0.020
Rel. FM = BCS_{Wi}	369.94	0.08	38.31	24.88	0.136
FM = BCS_M	389.20	0.10	98.45	59.10	0.108
FM = BCS_W	385.44	0.21	68.32	25.99	0.014
FM = BCS_F	381.86	0.31	89.98	26.65	0.002
FM = BCS_{Wi}	384.58	0.23	91.05	32.32	0.009
FFM = BCS_M	463.68	0.06	283.50	223.40	0.216
FFM = BCS_W	463.56	0.06	137.80	104.90	0.200
FFM = BCS_F	463.50	0.06	152.90	114.50	0.193
FFM = BCS_{Wi}	462.08	0.11	231.60	129.00	0.084
Mass = BCS_M	472.08	0.08	382.00	259.60	0.153
Mass = BCS_W	471.33	0.10	206.10	120.50	0.099
Mass = BCS_F	470.82	0.12	242.90	130.50	0.074
Mass = BCS_{Wi}	469.59	0.16	322.70	147.50	0.038

Rel. FM: Relative fat mass; FM: fat mass; FFM: fat free mass; BCS_M: BCS_{Morfeld}; BCS_W: BCS_{Wemmer}; BCS_F: BCS_{Fernando}; BCS_{Wi}: BCS_{Wijeyamohan}.

Table 4.

Estimates for each BCS system in statistical models to predict body composition, adjusted for sex and age.

Model	AIC	R ²	Estimate	SE	P
Rel. FM = BCS _M Sex Age	365.34	0.33	73.00	43.31	0.105
Rel. FM = BCS _W Sex Age	362.61	0.39	43.91	18.58	0.027
Rel. FM = BCS _F Sex Age	356.87	0.50	62.93	17.93	0.002
Rel. FM = BCS _{Wi} Sex Age	360.85	0.43	63.37	23.12	0.011
FM = BCS _M Sex Age	389.30	0.21	157.57	66.44	0.026
FM = BCS _W Sex Age	383.30	0.37	95.79	26.88	0.002
FM = BCS _F Sex Age	378.64	0.46	116.35	26.45	<0.001
FM = BCS _{Wi} Sex Age	381.96	0.40	128.38	33.72	<0.001
FFM = BCS _M Sex Age	444.73	0.589	359.84	178.76	0.055
FFM = BCS _W Sex Age	440.27	0.65	221.62	74.34	0.006
FFM = BCS _F Sex Age	442.39	0.62	210.46	82.56	0.018
FFM = BCS _{Wi} Sex Age	441.34	0.63	269.42	97.35	0.011
Mass = BCS _M Sex Age	458.08	0.52	517.42	226.90	0.032
Mass = BCS _W Sex Age	452.34	0.61	317.42	92.21	0.002
Mass = BCS _F Sex Age	453.33	0.59	326.83	100.38	0.003
Mass = BCS _{Wi} Sex Age	453.00	0.60	397.82	119.88	0.003

Rel. FM: Relative fat mass; FM: fat mass; FFM: fat free mass; BCS_M: BCS_{Morfeld}; BCS_W: BCS_{Wemmer}; BCS_F: BCS_{Fernando}; BCS_{Wi}: BCS_{Wijeyamohan}.

Table 5.

Estimates for each BCS system in statistical models to predict body composition, adjusted for sex, age, zoo, and familial relatedness.

Model	AIC	Estimate	SE	P
Rel. FM = BCS_M Sex Age Zoo Pedigree	271.98	111.98	45.97	0.015
Rel. FM = BCS_W Sex Age Zoo Pedigree	265.29	86.42	19.45	<0.001
Rel. FM = BCS_F Sex Age Zoo Pedigree	265.48	81.32	18.38	<0.001
Rel. FM = BCS_{Wi} Sex Age Zoo Pedigree	263.68	117.09	24.97	<0.001
FM = BCS_M Sex Age Zoo Pedigree	283.59	208.08	68.66	0.002
FM = BCS_W Sex Age Zoo Pedigree	275.54	144.48	27.43	<0.001
FM = BCS_F Sex Age Zoo Pedigree	281.40	115.75	29.57	<0.001
FM = BCS_{Wi} Sex Age Zoo Pedigree	277.72	193.53	42.28	<0.001
FFM = BCS_M Sex Age Zoo Pedigree	328.91	260.50	227.29	0.252
FFM = BCS_W Sex Age Zoo Pedigree	327.87	221.85	110.66	0.045
FFM = BCS_F Sex Age Zoo Pedigree	329.73	155.28	112.20	0.166
FFM = BCS_{Wi} Sex Age Zoo Pedigree	328.99	219.87	153.17	0.151
Mass = BCS_M Sex Age Zoo Pedigree	336.26	457.56	292.17	0.117
Mass = BCS_W Sex Age Zoo Pedigree	334.23	367.67	132.48	0.006
Mass = BCS_F Sex Age Zoo Pedigree	335.88	312.45	134.28	0.020
Mass = BCS_{Wi} Sex Age Zoo Pedigree	335.81	397.54	200.63	0.048

Rel. FM: Relative fat mass; FM: fat mass; FFM: fat free mass; BCS_M: BCS_Morfeld; BCS_W: BCS_Wemmer; BCS_F: BCS_Fernando; BCS_{Wi}: BCS_{Wi}jeyamohan; Pedigree: familial relatedness.