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RESEARCH ARTICLE

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Development of an index based on ultrasonographic measurements for the objective appraisal of body condition in Andalusian horses

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Abstract

Body condition scoring (BCS) is an indirect measure of the level of subcutaneous fat; however, by measuring the subcutaneous fat thicknesses (SFT), the precision of the degree of fatness assessment is improved. The aims were: 1) to develop an alternative body fat scoring index (BFSI) based on ultrasonographic measurements; 2) to assess the agreement between BCS and the new index applied to Andalusian horses; 3) to adjust the BCS cut-off values (if necessary) for overweight and obesity in this breed. One hundred and sixty-six Andalusian horses were included in this cross sectional study. On each horse, BCS, body fat percentage (BF%) and ultrasonography of SFT at localized deposits were evaluated. According to BFSI five possible body categories were established. Only one horse (0.6%) was classified as emaciated, 9.0% as thin, 74.7% as normal, 11.4% as overweight and 4.2% as obese. Despite higher BCS and SFT values were observed compared to other breeds, most of the horses evaluated presented a normal body condition under the new BFSI. BCS and BFSI were significantly associated (p<0.001), however, the concordance was low (weighted Cohen's kappa coefficient, 0.262 ± 0.071; p=0.004). Using BFSI, obese horses had significantly greater BF% than the rest of categories (p<0.001). BCS showed a good diagnostic accuracy for detection overweight (AUC = 0.759 ± 0.055; p<0.001) and obese (AUC = 0.878 ± 0.050; p=0.001) horses; redefining the cut-off values for overweight and obesity condition as 7.5/9 and 8.5/9 respectively in Andalusian horses.

Additional keywords: ultrasonography; subcutaneous fat deposits; objective score; adiposity; obesity; index.

Abbreviations used: AUC (area under the curve); BMI (body mass index); BCS (body condition score); BFSI (body fat scoring index); ROC (receiver operating characteristic curve); SFT (subcutaneous fat thickness); SFT-N25% (SFT over the first third of the neck-length); SFT-N50% (SFT over the second third of the neck-length); SFT-N75% (SFT over the last third of the neck-length); SFT-S (SFT behind the shoulder); SFT-Rb (SFT over the ribs); SFT-R (SFT over the rump); SFT-TH (SFT over the tailhead); BF% (body fat percentage).

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Introduction

Body mass index (BMI) is the most common objective measure used to classify excess adiposity in human beings (Lau *et al.*, 2007). Although BMI has also been applied in horses and ponies (Donaldson *et al.*, 2004; Carter *et al.*, 2009a; Thatcher *et al.*, 2012; Banse & McFarlane, 2014), the main system to assess body condition in horses is based on assigning a subjective body condition score (BCS). This method consists in evaluating the deposition of subcutaneous fat in specific body regions and the subsequent assignment of a score considering established criteria through a palpation and visual assessment (Carter & Dugdale, 2013). Even though it has become a universally accepted method to estimate the degree of fatness (Dugdale *et al.*, 2012), BCS possesses well-known limitations at the individual level as occurs with the BMI, including the inability of both systems to directly distinguish between lean and fat tissue (Frankenfield *et al.*, 2001; Geor & Harris, 2013). Therefore, with the same BCS, substantial variation in adiposity can occur (Dugdale *et al.*, 2011a). Furthermore, in the case of BCS, its inherent subjectivity and, thus, the semi-quantitative nature of this evaluation, lead to the belief that these scoring systems are unreliable and are necessary clinically more applicable and useful subdivisions to differentiate horses with higher scores (Burkholder, 2000; Dugdale *et al.*, 2012).

Considering that the limitations are common not only in horses but also in all animal species where BCS has been utilized, several studies have developed alternative methods to differentiate body condition using objective criteria (Bewley *et al.*, 2008; Azzaro *et al.*, 2011; Halachmi *et al.*, 2013; Das & Paksoy, 2015; Pfeifer *et al.*, 2017). However, some of them require specialised equipment, software and personnel to interpret the results making them less applicable in clinical settings. Moreover, most of them have been implemented only for their use in cattle (Bewley *et al.*, 2008; Azzaro *et al.*, 2011; Halachmi *et al.*, 2013; Das & Paksoy, 2015; Pfeifer *et al.*, 2017) and therefore, the necessity to find a suitable method to objectively evaluate the body condition in horses is still lacking. Against this perspective, taking into account that body weight alone is not a good indicator of relative adiposity (Carter & Dugdale, 2013), a good assessment of body fat reserves, minimizing the influence of body dimensions and intestinal contents, can be achieved by evaluating ultrasonographically the amount of subcutaneous fat.

Therefore, the aims of this study were: 1) to develop an alternative body scoring index based on the ultrasonographic evaluation of localized subcutaneous fat deposits; 2) to assess the agreement between BCS and the new index applied to Andalusian horses; 3) to adjust the BCS cut-off values (if necessary) for overweight and obesity in this breed.

Methods

Animals

From a population of over 1,500 Andalusian horses located in south-eastern Spain (comprising the provinces of Albacete, Alicante and Murcia), 166 (6.7 ± 3.7 years, 78 stallions and 88 females) were utilized in this cross sectional study for the development of a new body fat scoring index (BFSI) built on the application of ultrasonographic measurements. The sample was randomly selected in order to ensure a sufficient number of horses from both genders representing their score characteristics, and hence testing the reliability of this method.

Body and fat measurements

Using the nine point scale described by Henneke *et al.* (1983), two independent and trained evaluators assigned the scores on each horse and the average value was used as final BCS. Considering previous descriptions (Thatcher *et al.*, 2012) but also the phenotypic characteristics of the Andalusian horses, the following four body categories were established: thin horses (if BCS ≤ 4.5), normal body condition (if BCS 5- 6.5), overweight (if BCS 7-7.5) and obese horses (if BCS ≥ 8).

Afterwards, the amount of fat reserves were measured evaluating the subcutaneous fat thickness (SFT) by real-time ultrasound at seven localized fat deposits which included: three equidistant areas along the neck crest (SFT-N25%, SFT-N50%, SFT-N75%), behind the shoulder (SFT-S), the ribs (SFT-Rb), the rump (SFT-R) and the tailhead (SFT-TH) as previously described (Martin-Gimenez *et al.*, 2016). Ultrasonography was carried out using a portable Honda Electronics HS-1500V (Aichi, Japan) ultrasound device in B-mode with a linear transducer at 7.5 MHz frequency. BCS evaluation and the ultrasonographic measurements were taken for each horse on the same day. Measurements were obtained by freezing the image on the screen and measuring the position of maximal fat thickness. All measurements of SFT were performed in triplicate by the same researcher. To assess the reliability of repeated measurements, the intraclass correlation coefficients were calculated showing a significant repeatability ($p<0.001$) of 96.8% for SFT-N25%, 95.6% for SFT-N50%, 97.2% for SFT-N75%, 98.6% for SFT-S, 98.6% for SFT-Rb, 99.4% for SFT-R and 99.6% for SFT-TH. Because the agreement between different measurements was good (Fleiss, 1986) mean values of the three measurements were used for statistical analyses.

Body fat percentage (BF%) was also calculated to assess the new BFSI as monitor parameter of body fat stores. This variable was estimated from the equation of Kane *et al.* (1987) where: BF% = 2.47 + 5.47 * (rump fat in cm). The site to measure rump fat was determined by placing the probe over the rump at approximately 5 cm lateral from the midline at the centre of the pelvic bone (Westervelt *et al.*, 1976).

All the measurements and body condition estimations were collected following informed consent from the owners.

Statistical analysis

Data were expressed as mean ± standard deviation (SD) or percentage, as appropriate. Normality of quantitative variables was checked using the Kolmogorov-Smirnov test.

To test the usefulness of SFT measurement technique, two different approaches were performed: 1) analysis of variance (ANOVA) to determine the relationship among the different scores included in the BCS system and the SFT values and; 2) association between BCS, BF% and SFTs using the Pearson's and Spearman's correlation coefficients. Besides, Student's t test for independent samples was used to evaluate the association among the gender and the SFTs (Daniel, 2000).

Construction of the new body fat scoring index

Mean and SD of each SFT measurement were calculated. Depending on the number of SDs from the average SFT value, a standardized score (equal to the integer part of the standardized residual) was assigned to every SFT on each horse. The difference between an individual SFT value and the mean divided by the SD corresponds to the standardized residual (Daniel, 2000). To simplify these calculations and taking into account the mean and SD, different intervals were established attributing to them the following standardized scores: -2 (- ∞, mean – 2*SD], -1 (mean – 2*SD, mean – SD], 0 (mean – SD, mean + SD]), +1 [mean + SD, mean + 2*SD), +2 [mean + 2*SD, mean + 3*SD) and +3 [mean + 3*SD, + ∞). The overall objective score of a horse resulted from the sum of all scores obtained at each anatomical area. Differences between genders in overall objective scores were assessed using Student's t test or Mann-Whitney test depending on normality. Mean and SD of the overall objective scores and their intervals, similarly to what was done with each of the SFT measurements, were estimated defining five possible BFSI categories. To analyse the association between BCS and the BFSI, Pearson's Chi-square test, Spearman's correlation and Cohen's kappa coefficients were calculated (Cohen, 1968; Daniel, 2000; Thrusfield, 2005).

Spearman's correlations were used to evaluate the association between BF%, BCS and BFSI. To determine changes in BF% across the BCS and BFSI categories, an ANOVA was performed. Duncan *post hoc* test was used to separate between significant means.

The reliability of the BCS to distinguish between horses that did and did not exhibit an overweight or obesity state defined by the new BFSI, was estimated by calculating the area under the curve (AUC). The coordinates of the receiver operating characteristic curve (ROC) were used to set the cut-off values that maximised the accuracy (proportion of true results) (Greiner *et al.*, 2000). Their confidence intervals (CI) were calculated using the Wilson's score method (Wilson, 1927).

The analyses were carried out with the statistical software program IBM SPSS for Windows Vers. 19, except for the calculation of weighted Cohen's kappa coefficient that was used StatsToDo (https://www.statstodo.com/CohenFleissKappa_Pgm.php). Values of p<0.05 were considered significant.

Results

The global mean of BCS was 6.12 ± 1.05 without significant differences between both genders (p=0.695). Agreement between the two body condition evaluators was moderate (Cohen's kappa weighted =0.493, $CI_{95\%}$: 0.423, 0.564; p<0.001). Mean values ± SD of the seven SFT measurements are described in Table 1. Regard to the gender, males presented significantly

Table 1. Global mean ± SD values of subcutaneous fat thickness (SFT) and their relationship with gender, body condition score (BCS) and body fat percentage (BF%).

Measurement[1]	Overall (n = 166)	Differences between sexes			Relationship[2] with	
		Males (n = 78)	Females (n = 88)	$p_{\text{Student's t}}$	BCS	BF%
SFT-N25%	6.65 ± 1.94	7.48 ± 1.88	5.91 ± 1.68	<0.001	0.143 (0.066)	0.072 (0.354)
SFT-N50%	11.22 ± 2.82	11.93 ± 2.81	10.58 ± 2.68	0.002	0.135 (0.083)	0.120 (0.125)
SFT-N75%	10.08 ± 3.41	10.24 ± 3.03	9.94 ± 3.72	0.576	0.267 (0.001)	0.287 (<0.001)
SFT-S	7.24 ± 2.93	7.80 ± 3.13	6.74 ± 2.66	0.020	0.204 (0.008)	0.242 (0.002)
SFT-Rb	7.44 ± 2.40	8.02 ± 2.75	6.93 ± 1.92	0.003	0.326 (<0.001)	0.238 (0.002)
SFT-R	14.43 ± 5.38	13.75 ± 4.67	15.03 ± 5.90	0.126	0.491 (<0.001)	1.000 (<0.001)
SFT-TH	23.72 ± 8.74	22.38 ± 7.41	24.92 ± 9.65	0.062	0.611 (<0.001)	0.626 (<0.001)

[1]SFT-N25%, SFT over the first third of the neck length; SFT-N50%, SFT over the second third of the neck length, SFT-N75%, SFT over the last third of the neck length; SFT-S, SFT behind the shoulder; SFT-Rb, SFT over the ribs; SFT-R, SFT over the rump; SFT-TH, SFT over the tailhead area. [2]Correlation coefficients and p-values in parenthesis

higher values at four SFT measurements (SFT-N25%, SFT-N50%, SFT-S and SFT-Rb) than females (Table 1). Subcutaneous fat thicknesses (with the exception of SFT-N25% and SFT-N50%) were significantly correlated with BCS and BF% (Table 1). Likewise, most of SFT measurements were significantly different depending on the BCSs assigned in the sample (Table 1), increasing their values as the BCS does, despite the low correlations (Fig. 1).

Due to significant differences in SFT values were observed according to the gender, firstly different sets of intervals had to be created stratified by sex to define the standardized scores corresponding to each fat deposit (Table 2). Secondly, the overall objective score of a horse was calculated adding up the seven standardized scores obtained. Global mean of the new BFSI was 0.265 ± 2.697 being similar between males and females (*p*=0.910). Based on the absence of differences between genders after standardizing the scores, it was possible to define the final scores common to both genders. In this instance, five body categories (emaciated, thin, normal, overweight and obese) were proposed so that an animal with an overall standardized score equal to or greater than 6 was considered as obese, while an animal with a score between 3 and 5 (both inclusive) was classified as overweight (Table 3).

The application of the BCS system showed that the majority of the samples were distributed among the interval scores of 5 and 6.5 (63.9%) and 7-7.5 (22.9%). In addition, 8.4% and 4.8% were classified as thin horses (BCS ≤ 4.5) and obese (BCS ≥ 8) respectively. Concerning the BFSI, only one horse was classified as emaciated (0.6%) (because of its low representativeness, it was excluded for further calculation of agreement). Most of the horses (74.7%) had a normal body condition and 11.4% were overweight. Fifteen horses (9.0%) were considered as thin horses while seven (4.2%) were categorized as obese (Table 4 & Fig. 2).

The correlation between BCS and the BFSI was significant and moderate attending to the Spearman's correlation coefficient value (rho=0.428; *p*<0.001). In addition, the association among the BCS and the BFSI categories was highly significant (*p*<0.001), however the concordance between both body scoring methods was very low, evidenced by a weighted Cohen's kappa coefficient of 0.262 ± 0.071 (*p*=0.004) (Table 4). In this manner, it could be observed as remarkable results, that half of horses with BCS ≥ 8 were classified as having a normal body condition with the BFSI, while the remaining 50% were equally distributed among overweight and obese. Similarly, in the case of the horses with BCS = 7-7.5, more than half (65.8%) were categorized as normal with the BFSI, 23.7% as overweight and 10.5% as obese (Table 4).

The global BF% was 10.36 ± 2.94 without significant differences between both genders (*p*=0.126). Otherwise, BF% was significantly correlated with BCS (rho=0.491; *p*<0.001) and BFSI (rho=0.503; *p*<0.001). Depending on the BCS categorization, significant differences (*p*<0.001) were found among thin (7.74 ± 2.89) *vs* normal horses (9.82 ± 2.18) and, these two categories *vs* overweight (12.10 ± 2.73) and obese (13.84 ± 5.42). However, no differences were observed between overweight and obese horses. On the contrary when BFSI was applied, obese horses (16.03 ± 4.45) presented higher BF% values (*p*<0.001) respect to overweight horses (12.50 ± 2.78), as well as these two categories respect to normal (9.93 ± 2.36) and thin

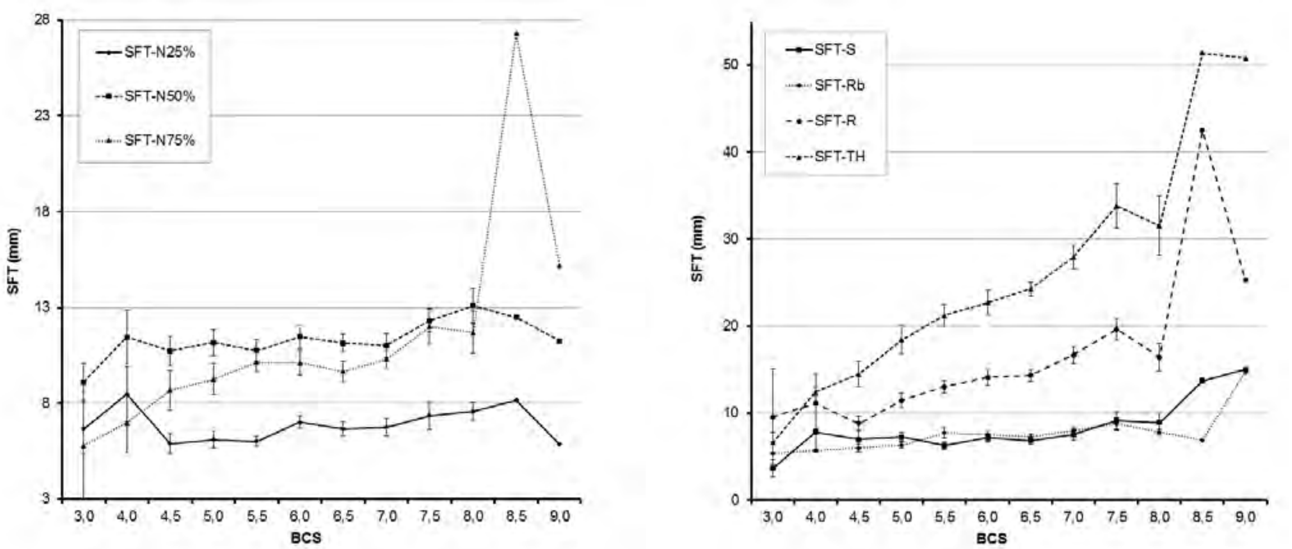

Figure 1. Variation of subcutaneous fat thickness (SFT) (mean ± standard error) across the body condition scores (BCS). Significance of ANOVA: SFT-N25%, *p*=0.238; SFT-N50%, *p*=0.740; SFT-N75%, *p* <0.001; SFT-S, *p*=0.002; SFT-Rb, *p*=0.005; SFT-R, *p* <0.001; SFT-TH, *p* <0.001.

Table 2. Standardized scores and corresponding intervals for each subcutaneous fat thickness (SFT) measurement according to the gender

Measure[1]	Score	Males		Females	
		min	max	min	max
SFT-N25%	-2		3.721		2.545
	-1	3.721	5.602	2.545	4.226
	0	5.602	9.362	4.226	7.589
	+1	9.362	11.242	7.589	9.270
	+2	11.242	13.122	9.270	10.952
	+3	13.122		10.952	
SFT-N50%	-2		6.302		5.228
	-1	6.302	9.115	5.228	7.906
	0	9.115	14.742	7.906	13.262
	+1	14.742	17.555	13.262	15.939
	+2	17.555	20.368	15.939	18.617
	+3	20.368		18.617	
SFT-N75%	-2		4.166		2.500
	-1	4.166	7.201	2.500	6.220
	0	7.201	13.271	6.220	13.658
	+1	13.271	16.306	13.658	17.377
	+2	16.306	19.341	17.377	21.097
	+3	19.341		21.097	
SFT-S	-2		1.532		1.428
	-1	1.532	4.666	1.428	4.084
	0	4.666	10.933	4.084	9.398
	+1	10.933	14.067	9.398	12.055
	+2	14.067	17.201	12.055	14.712
	+3	17.201		14.712	
SFT-Rb	-2		2.529		3.094
	-1	2.529	5.275	3.094	5.014
	0	5.275	10.768	5.014	8.853
	+1	10.768	13.514	8.853	10.772
	+2	13.514	16.260	10.772	12.692
	+3	16.260		12.692	
SFT-R	-2		4.412		3.227
	-1	4.412	9.081	3.227	9.130
	0	9.081	18.420	9.130	20.935
	+1	18.420	23.089	20.935	26.838
	+2	23.089	27.758	26.838	32.741
	+3	27.758		32.741	
SFT-TH	-2		7.556		5.611
	-1	7.556	14.968	5.611	15.264
	0	14.968	29.792	15.264	34.570
	+1	29.792	37.204	34.570	44.223
	+2	37.204	44.617	44.223	53.876
	+3	44.617		53.876	

[1]SFT-N25%, SFT over the first third of the neck length; SFT-N50%, SFT over the second third of the neck length, SFT-N75%, SFT over the last third of the neck length; SFT-S, SFT behind the shoulder; SFT-Rb, SFT over the ribs; SFT-R, SFT over the rump; SFT-TH, SFT over the tailhead area.

Table 3. Body condition classification using the new body fat scoring index (BFSI)

Min	Max	BFSI	Categories
	-5.129	≤ -6	Emaciated
-5.129	-2.432	[-5, -3]	Thin
-2.432	2.962	[-2, 2]	Normal
2.962	5.659	[3, 5]	Overweight
5.659		≥ 6	Obese

horses (8.84 ± 2.71). In this case, BF% was similar between thin and normal horses.

Diagnostic accuracy of BCS to distinguish overweight or obese horses from all other horses was assessed by evaluation of areas under the ROC curves. For the first case, BCS had an AUC =0.759 ± 0.055 (p<0.001). In the second case, BCS presented an AUC = 0.878 ± 0.050 (p=0.001) (Fig. 3). The ROC curve analysis was also employed to determine the BCS cut-off values for detecting overweight (BFSI = [3, 5]) and obese (BFSI ≥6) horses, and these values were 7.5 with an accuracy of 85.54% ($CI_{95\%}$: 79.39%, 90.09%) and 8.5 with an accuracy of 96.99% ($CI_{95\%}$: 93.14%, 98.71%), respectively.

Discussion

Previous studies have proposed that Andalusian horses have an innate tendency towards obesity. However, previous appraisements of their body condition have been made based on palpation and visual estimation (Bamford *et al.*, 2013; Potter *et al.*, 2013). The main goals of this study have been to estimate objectively the body condition in this breed, and to demonstrate that the assumed BCS cut-off values indicative of overweight and obesity state need to be modified in this breed.

Body scoring systems have been applied across diverse animal species from its use in primates (Clingerman & Summers, 2005), wild animals (Gerhart *et al.*, 1996), cattle (Edmonson *et al.*, 1989) and

Table 4. Association among the body condition score (BCS) and the new body fat scoring index (BFSI) categories.

BCS			BFSI (%)				
Categories	n	%	Emaciated (≤-6)	Thin [-5, -3]	Normal [-2, 2]	Overweight [3, 5]	Obese (≥6)
Thin (BCS ≤4.5)	14	8.4	7.1	14.3	78.6	0.0	0.0
Normal (BCS = 5, 6.5)	106	63.9	0.0	12.3	79.2	7.5	0.9
Overweight (BCS = 7, 7.5)	38	22.9	0.0	0.0	65.8	23.7	10.5
Obese (BCS ≥8)	8	4.8	0.0	0.0	50.0	25.0	25.0
Total	166	100	0.6	9.0	74.7	11.4	4.2

Significance of Pearson's Chi-square test, p<0.001

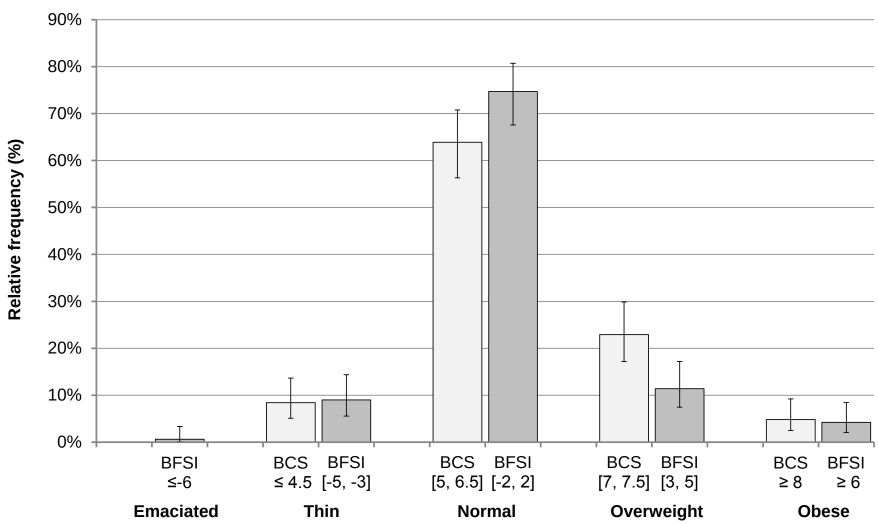

Figure 2. Distribution and comparison of body condition categories when body condition score (BCS) and body fat scoring index (BFSI) are applied.

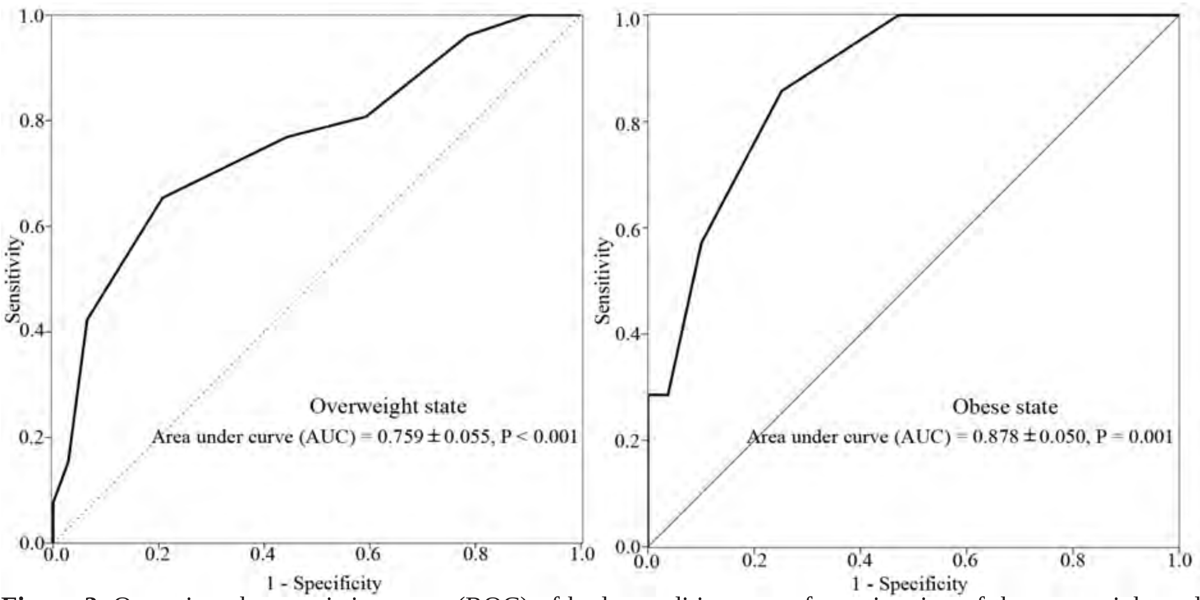

Figure 3. Operating characteristic curves (ROC) of body condition score for estimation of the overweight and obese states.

companion animals including horses (Henneke *et al.*, 1983; Carroll & Huntington, 1988; Laflamme, 1997; Mawby *et al.*, 2004). Otherwise, ultrasonography has demonstrated to be an accepted method for measuring fat reserves in farm species (Silva & Cadavez, 2012) and equids (Gentry *et al.*, 2004) due to its objectivity, repeatability of the technique (Martin-Gimenez *et al.*, 2016), low cost and the possibility of being used in field conditions (Quaresma *et al.*, 2013). Thus, in many species, ultrasonography has also been utilized to validate the condition scoring process (Domecq *et al.*, 1994; Gentry *et al.*, 2004; Alapati *et al.*, 2010; Morfeld *et al.*, 2014) and/or to predict the total fat content using mathematical equations that frequently include some SFT measurement (Westervelt *et al.*, 1976; Kane *et al.*, 1987; Stephenson *et al.*, 1998). Conversely, in this case the ultrasonography was used to create a new objective scoring system. For that, a body assessment method was built considering the objectivity provided by the ultrasonographic evaluation of subcutaneous fat deposits and keeping in mind that none of the SFT measurements by themselves have been able to result in a good prediction of BCS demonstrated by the low correlations observed. In relation with this, it is important to notice that despite the high SFT values registered, it was also observed a considerable heterogeneity in fat deposition patterns between individual animals that could explain these weak relationships. So that horses with high BCS can present certain areas with low amount of fat and vice versa. To the authors' this may be explained by two ways. Firstly, although subcutaneous adipose tissue is a key determinant of BCS, the subjectivity of this method make that it can be influenced by others factors, such as the conformation inherent to the breed evaluated. Then, higher scores may not be the consequence of high SFT. Secondly it has been shown that even in homogeneous populations the distribution of fat between different deposits is highly variable (Pond, 1998). Besides, although the order in which individual adipose tissues are recruited in the development of obesity, is getting to be understood in genetically modified species (Reed *et al.*, 2006), in horses the knowledge of body composition, control of fat deposition, and mobilisation warrants further investigation (Argo, 2009).

Fat deposits examined previously by ultrasonography in equids differed from study to study (Gentry *et al.*, 2004; Carter *et al.*, 2009b; Argo *et al.*, 2012) probably attributed to breed differences and to the ease of obtaining and reading ultrasonographic measurements. Also, if a specific ultrasound imaging analysis software could be developed to decrease the time-consuming and increase the precision of fat measuring, especially with those kind of protocols that involve taking the measurements in triplicate (to increase the accuracy), maybe less differences would be described in the literature. In the present study, the regional deposits selected were closely related to the anatomical locations on which the BCS system is measured and corresponding with the areas with greater tendency to accumulate fat in this breed. Comparing the results, is observed that the thicknesses of fat in most of the areas evaluated were greater than those reported in previous studies (Cartmill *et al.*, 2006; Dugdale *et al.*, 2011b; Quaresma *et al.*, 2013). In addition, due to the representativeness of the sample, the degree of deviation of each SFT measurement on each animal in relation to the average of the sample was estimated. In this manner, the degree of fatness on each body area could be rated and continue monitoring over time which is important since increased regional adiposity is a health issue in horses because its association with altered metabolic states (Johnson, 2002).

The low concordance observed between the BCS and the BFSI could be explained because, although the proportion of horses at the extremes of both scoring scales were relatively similar, the differences were evident in animals with intermediate scores where the proportion of horses classified as normal *vs* overweight varied clearly between both methods. Otherwise, it is worth mentioning that albeit the high SFTs registered and the mean BCS was greater compared to previous studies (Pratt-Phillips *et al.*, 2010; Turner *et al.*, 2011; Wagner & Tyler, 2011), the overall BF% was lower than in other breeds (Vick *et al*., 2007; Adams *et al*., 2009; Ragnarsson & Jansson, 2011). Considering these data, we determined that Andalusians were not as overweight as it could appear if we only use BCS to evaluate the body condition. This also agree with the fact that applying the BFSI, the majority of Andalusians (75.2%) presented a normal body condition, which should be considered in the average for this breed. These findings suggest that the subjective scoring underestimate the optimal body condition and overestimate the overweight state in this breed.

Regardless of the significant association among the BCS and BFSI established categories, it should be noted that some striking misjudgements were shown. Notable was that, among the horses with BCS 5-6.5, the BCS was not sensitive enough to detect those horses that would better fit in the overweight category under the new BFSI. Likewise, most of the horses with BCS ≥ 7 would present a normal body condition using the BFSI, suggesting again that the subjective scoring method overestimates the overweight and obesity states in Andalusian horses and supporting the need to adjust the BCS ranges in accordance with breed specific criteria.

Among the available methodologies to quantify objectively the body fat content in live horses (Kearns *et al.*, 2002a), estimation of BF% using the method developed by Westervelt *et al.* (1976) and Kane *et al.* (1987) suppose the most feasible, cost-effective and prevalent reported method (Kearns *et al.*, 2001, 2002b, 2006; Vick *et al.*, 2007; Adams *et al.*, 2009; Ragnarsson & Jansson, 2011). For this reason, BF% was estimated in this study as a quantitative method of total fat mass assessment and hence, as validation variable to corroborate de adiposity level of each body condition category. The degree of correlation between the BF% and BCS was lower than previously described (Henneke *et al.*, 1983; Vick *et al.*, 2007). These discrepancies could be explained because unlike other studies, in this case both genders have been considered and the number of animals included was much higher. Nevertheless, the use of BFSI improved the degree of association with the BF% and showed a greater sensitivity to distinguish between overweight and obese individuals which support the reliability and potential application of this system by clinicians to detect the subgroups at greater risk of metabolic disturbances.

Undoubtedly, modern nutritional and management practices are contributing to the increase in equine obesity prevalence across most of breeds (Scheibe & Streich, 2003), however many times the scales applied (Henneke *et al.*, 1983; Kohnke, 1992; Kienzle & Schramme, 2004) and terminology to classify the body condition vary making not comparable the results among studies. In relation with this and the importance of establishing specific criteria to define the overweight and obesity, the AUCs showed that the BCS has a good diagnostic accuracy (Greiner *et al.*, 2000). However, considering the faithful fulfilment of the original scale described by Henneke *et al.* (1983), the results confirm that the scores to designate these two body categories (obese and overweight) should be raised at least in Andalusian horses. Previous studies in which the conventional body scoring system has been utilized, obese horses have been described using different cut-off values (Gentry *et al.*, 2002; Hoffman *et al.*, 2003; Gentry *et al.*, 2004; Buff *et al.*, 2006; Frank *et al.*, 2006; Vick *et al.*, 2006; Waller *et al.*, 2006; Ungru *et al.*, 2012). This lack of consensus among different researches to define obesity stands out the relevance of these results where based strictly on an objective appraisal of the body condition (SFT ultrasonography) and in accordance with a quantitative corroborated obesity variable (%BF) it has been possible to fix concrete cut-off values adjusted to a specific breed.

The new body scoring method presents as main advantages its objectivity, non-invasive nature, quickness, safety, easiness to perform and applicability on a variety of subject populations. Additionally, body assessments by this method can be repeated over an unlimited period of time, making longitudinal studies realizable. However, it should be taken into account that in Spain the castration of horses from this breed is not frequent. This was reflected in the sample studied where all selected males were ungelded. This could be considered as a limitation since the usefulness of BFSI over gelding horses has not been possible to verify, being necessary further investigations to test its application in these horses as well as its repeatability across different breeds.

In conclusion, the majority of Andalusian horses evaluated in this study presented a body condition, which could be considered in the average for this breed. The developed BFSI suggests that the subjective assessment of body condition by conventional BCS systems overestimates the degree of fatness in these horses. In addition, this system discusses the cut-off values traditionally established in BCS scale to define the overweight and obesity and, indicates that it would be necessary to increase them by at least 0.5 points in Andalusians to detect correctly those horses with excess of adiposity.

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