

Beyond scoring systems: usefulness of morphometry considering demographic variables, to evaluate neck and overall obesity in Andalusian horses

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Morphometry has proven to be a useful tool, both for the clinician and horse owners, for evaluating the body condition in equids due to its objectivity, easiness and capacity for detection of important metabolic disturbances. However, limited information is available on the use of morphometric ratios to characterize regional and overall adiposity and much less about their application in different genders, ages and horses with different levels of obesity. The objectives were to evaluate body and neck absolute measurements and ratios; factors affecting them such as the influence of gender, age, appearance of the neck crest and overall body condition and; relationships among these measurements. A total of 154 Andalusian horses classified according to their gender, age, body score status and cresty neck condition were evaluated in this cross-sectional study. Two evaluators assigned a body condition score (BCS, 1 to 9) and a cresty neck score (CNS, 0 to 5) to each horse. Horses were divided into males and females; young (2 to 5 years) and adults (6 to 15 years); obese (BCS ≥ 7) and non-obese (BCS < 7); cresty neck (CNS ≥ 3) and non-cresty neck horses (CNS < 3). Morphometric measurements (cm) included were: height at the withers (HW); body length (BL), girth (GC) and waist (WC) circumferences; neck length (NL); three neck circumferences (NCs), over the first (NC_{25%}), the second (NC_{50%}) and the third part (NC_{75%}) of the NL and neck crest height (NCH). These measurements were also used to calculate the following ratios: GC:HW, WC:HW, GC:BL, WC:BL, NC_{25%}:HW, NC_{50%}:HW, NC_{75%}:HW, NC_{25%}:BL, NC_{50%}:BL, NC_{75%}:BL, NC_{25%}:NL, NC_{50%}:NL, NC_{75%}:NL, NC_{25%}:NCH, NC_{50%}:NCH and NC_{75%}:NCH. The results showed that most of the absolute measurements and ratios were greater than those described in other light breeds. In addition, most neck ratios were higher ($P < 0.050$) in males than in females, however, all body ratios were greater ($P < 0.001$) in females. Among the absolute measurements, WC in obese horses and NC_{25%} and NC_{75%} in cresty neck horses highlighted as higher. Either GC:HW or WC:HW and NC_{75%}:BL were alternative surrogates for the appraisal of overall and regional adiposity in Andalusians. Several interactions were observed between the gender and adiposity scoring systems affecting the morphometric evaluation. This study establishes absolute morphometric measurements and ratios in Andalusian horses. It also highlights the variability of morphometric values and how the outcome of these can be influenced by demographic variables and the breed analyzed. Further studies are necessary to set morphometric reference values in other breeds.

Keywords: morphometric measurements, morphometric ratios, body condition, regional adiposity, cresty neck

Implications

Among important disturbances, such as decreased athletic ability, orthopaedic disorders and poor thermo-regulation, obesity can lead to other health issues including insulin resistance and Equine Metabolic Syndrome, both of which can lead to laminitis. Andalusian horses are considered an easy keeper breed, and their tendency towards obesity together with their characteristic phenotype, make it necessary to propose

objective measurements to accurately assess the deposition of adipose tissue beyond the subjective scoring systems. Our results show that fat deposition patterns and body shape according to gender and age, influence the direct comparison among the several morphometric measurements created.

Introduction

Obesity is an emergent problem in domestic horses and pony populations that has health and welfare implications (Giles *et al.*, 2014). In large-scale human populations, anthropometric

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approaches based on simple clinical measures have been applied to indirectly estimate the body fat content and have demonstrated its usefulness in the prediction of obesity-related metabolic risk (Bosy-Westphal *et al.*, 2006). In the case of equids, there is no consensual definition of obesity and the most widely used indexes for evaluation and categorization of being obese or overweight are the body condition scoring systems (Henneke *et al.*, 1983; Carroll and Huntington, 1988). However, these methods may be less sensitive to detect variations in body fat distribution (e.g. regional adiposity), observe variations in adiposity over time and also it has been reported that owners cannot accurately estimate the body condition of their horses using these systems (Wyse *et al.*, 2008; Mottet *et al.*, 2009; Thatcher *et al.*, 2012). On the other hand, some studies have demonstrated the clinical applicability of morphometric measurements to estimate adiposity in horses (Frank *et al.*, 2006; Carter *et al.*, 2009a; Dugdale *et al.*, 2010) and distinct advantages over subjective evaluation scores. They provide objectivity, do not need previously trained examiners and are able to detect small variations in body condition (Dugdale *et al.*, 2011b) allowing a precise quantification when corrections in nutritional management are applied to horses (Dugdale *et al.*, 2010; Argo *et al.*, 2012). Similarly to what happens in human medicine, in horses with laminitis, some morphometric measurements have been used as prognostic indicators (Carter *et al.*, 2009b) due to their proven relationship with metabolic derangements (Frank *et al.*, 2006; Carter *et al.*, 2009b). Morphometric measurements may not reflect the same degree of adiposity due to differences in breed-type morphology (Brooks *et al.*, 2010; Catalano *et al.*, 2016). Moreover, there is a very probable influence of intrinsic variables such as the gender and age. Considering these facts, the aims of the present investigation were: (1) to study different body and neck morphometric measurements to determine the characteristics of these in Andalusian horses; (2) to evaluate the effects of gender and age factors that could affect morphometry and; (3) to clarify the relationships among these measurements and general and regional adiposity to identify which are more reliable to assess body condition in this breed.

Material and methods

Design of the study and sample size

Morphometric measurements on 154 Andalusian horses, including both genders with an age from 2 to 15 years, were collected in this cross-sectional study carried out across three geographic regions of South-East of Spain in 2012. All included animals were healthy based on the results of physical examination, complete blood count, and serum biochemical analysis. Due to the fact that the majority of weight gain occurs through the second trimester of gestation (Lawrence *et al.*, 1992), pregnant mares (>100 days of gestation) and also lactating mares were excluded in order to avoid confounding factors in the body condition and morphometric evaluation. To evaluate the effect of age on the morphometric measurements, the horses were grouped by age in two groups distinguishing between young horses

(2 to 5 years) and adult horses (6 to 15 years), because most of the horses reach their full dimensions at 5 years of age (Čoudková *et al.*, 2016). The distribution of horses concerning sex and age is shown in Table 1.

Owners or the person in charge of each horse provided an informed consent form for the participation in the study.

Body condition and cresty neck scoring

Two independent evaluators assessed the body condition score (BCS) (Henneke *et al.*, 1983) and cresty neck score (CNS) (Carter *et al.*, 2009a) based on a scale of 1 to 9 for BCS and 0 to 5 for CNS. Average scores of both evaluators were used for data analysis. Horses with a BCS $\geq 7/9$ were considered obese (Giles *et al.*, 2014) and horses with a CNS $\geq 3/5$ were considered cresty neck horses (Carter *et al.*, 2009a).

Morphometric measurements

In all, 25 morphometric variables were determined, including four from the body, five from the neck (Figure 1) and 16 from the body and neck crest ratios.

Morphometrics taken over the trunk included: (1) height at the withers (HW), the distance from highest point of withers to the ground (Brooks *et al.*, 2010); (2) body length (BL), from the point of the shoulder (intermediate tubercle of the

Table 1 Distribution of studied horses according to sex and age

	Age 2 to 5 years	6 to 10 years	P-value
Stallions	40 (51.3%)	38 (48.7%)	0.747
Mares	37 (48.7%)	39 (51.3%)	

Significance of Pearson's χ^2 test.

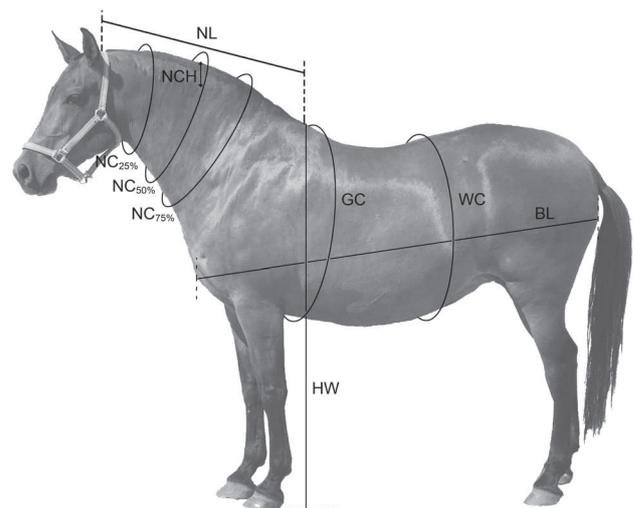


Figure 1 Morphometric measurement sites used in body condition and neck adiposity assessment, with the horse in an appropriate square stance and the neck held in a relaxed upright position at approximately a 45° angle from the horizontal. HW = wither height; BL = body length; GC = girth circumference; WC = waist circumference; NL = neck length; NC = neck circumference; NCH = neck crest height; NC_{25%} = neck circumference at 25% of neck length; NC_{50%} = neck circumference at 50% of neck length; NC_{75%} = neck circumference at 75% of neck length.

humerus) to a line perpendicular to the point of the buttock (ipsilateral ischiatic tuberosity) (Martinson *et al.*, 2014); (3) girth circumference (GC), surrounding the thoracic perimeter immediately behind the slope of the withers (at the third thoracic vertebra), caudal to the elbow (olecranon tuber) of both forelimbs and at the time of exhalation, so that the pulmonary volume is not considered (Carter *et al.*, 2009a); (4) waist circumference (WC), around the abdomen coinciding with the point of the umbilicus at two-thirds of the distance from the point of the shoulder to the point of the hip (tuber coxae) and at the end of the expiratory pause (Dugdale *et al.*, 2010).

In the neck area, neck length (NL) was measured as a straight line from the poll to the dorsal aspect of the withers (Frank *et al.*, 2006). Three neck circumferences (NCs) were evaluated perpendicular to this line. First one, halfway between the poll and the mid-neck length (NC_{25%}), the second one at the midway point between the poll and the dorsal aspect of the withers (NC_{50%}) and the last one halfway between the mid-neck length and the dorsal aspect of the withers (NC_{75%}) (Frank *et al.*, 2006). Neck crest height (NCH) was measured over the mid-neck length from the dorsal midline to the interface between the crest and the neck musculature (Carter *et al.*, 2009a). With these measurements, the following ratios were calculated: GC : HW, WC : HW, GC : BL, WC : BL, NC_{25%} : HW, NC_{50%} : HW, NC_{75%} : HW, NC_{25%} : BL, NC_{50%} : BL, NC_{75%} : BL, NC_{25%} : NL, NC_{50%} : NL, NC_{75%} : NL, NC_{25%} : NCH, NC_{50%} : NCH and NC_{75%} : NCH.

To ensure the continuity in the placement of measuring tools, all measurements (cm) were taken by one of the evaluators (always the same) in triplicate and before daily feeding. The measurements were taken from the left side of the horse while it was standing on flat ground in the right position with parallel limbs and neck held in a relaxed upright position at approximately a 45° angle from the horizontal. Neck and trunk circumferences, NCH and NL were measured with a plastified measuring tape (Kruuse, Langeskov, Denmark) and to measure the HW and the BL, a hipometer (specific measuring stick for large animals) (Hauptner, Dietlikon, Switzerland) was employed.

Intraclass correlation coefficients for the trunk and neck morphometric measurements were: 99.5% for HW, 99.4% for BL, 99.2% for GC, 99.0% for WC, 99.3% for NL, 99.2% for NCH, 98.4% for NC_{25%}, 98.2% for NC_{50%} and 98.4%

for NC_{75%}. Because the repeatability between different measurements was good (Fleiss, 1986), mean values of the three measurements were used for statistical analyses.

Statistical analysis

Quantitative variables were reported using mean \pm SD, CV and range (minimum and maximum). The association between two qualitative variables was checked using Pearson's χ^2 test. A GLM for each morphometric measurement and ratio (reported using estimated marginal means \pm residual errors) was applied using sex, age group, obesity status (BCS \geq 7) and cresty neck condition (CNS \geq 3) as fixed effects and also their paired interactions. Statistical analyses were carried out in SPSS 19.0 for Windows (IBM Corp., Armonk, NY, USA). A weighted Cohen's κ statistic indicating level of agreement was performed using StatsToDo (available at https://www.statstodo.com/CohenFleissKappa_Pgm.php). The α error was set at 0.05 and the power was set at 0.80.

Results

Description of studied parameters

Data were collected from 154 Andalusian horses: 78 stallions (50.6%) and 76 mares (49.4%) with an average age of 6.60 ± 3.72 years. The BCS ranged from 3 to 9 with an average of 6.07 ± 1.03 . Agreement between the two body condition evaluators was moderate (Cohen's κ weighted = 0.481, CI_{95%}: 0.404, 0.557; $P < 0.001$). In the case of CNS the range was from 1 to 5 with an average of 3.11 ± 0.80 . Agreement between the two cresty neck evaluators was substantial (Cohen's κ weighted = 0.691, CI_{95%}: 0.611, 0.770; $P < 0.001$). In all, 26% of the horses were considered to be obese and more than half (69.5%) were classified as cresty neck horses.

The distribution of horses concerning sex, age, BCS and CNS is shown in Table 2. When the association was analyzed among the fixed factors, it was found that some pairs were independent: sex *v.* age group ($P = 0.747$), sex *v.* obesity status ($P = 0.786$), age group *v.* obesity status ($P = 0.066$) and age group *v.* cresty neck condition ($P = 0.221$). However, sex and cresty neck condition were associated ($P < 0.001$) with 88.5% of stallions having cresty neck *v.* 50.0% of mares. Finally, obesity status and cresty neck condition were highly associated ($P < 0.001$), and cresty neck condition

Table 2 Distribution of studied horses (n = 154) according to sex, age, obesity status and cresty neck condition

	BCS <7	BCS \geq 7	P-value	CNS <3	CNS \geq 3	P-value
Stallions	57 (73.1%)	21 (26.9%)	0.786	9 (11.5%)	69 (88.5%)	<0.001
Mares	57 (75.0%)	19 (25.0%)		38 (50.0%)	38 (50.0%)	
2 to 5 years	62 (80.5%)	15 (19.5%)	0.066	27 (35.1%)	50 (64.9%)	0.221
6 to 10 years	52 (67.5%)	25 (32.5%)		20 (26.0%)	57 (74.0%)	
BCS <7				46 (40.4%)	68 (59.6%)	<0.001
BCS \geq 7				1 (2.5%)	39 (97.5%)	

BCS = body condition score; CNS = cresty neck score.
Significance of Pearson's χ^2 test.

Table 3 Description of neck and trunk morphometric measurements (cm) and ratios calculated in the sample (n = 154)

Variable	Mean ± SD	CV (%)	Range	
			Min	Max
Absolute measurements				
HW (cm)	157.8 ± 4.9	3.1	148.0	172.0
BL (cm)	159.4 ± 5.9	3.7	140.7	176.0
GC (cm)	191.3 ± 8.8	4.6	171.0	217.3
WC (cm)	197.2 ± 11.3	5.7	168.7	231.0
NL (cm)	88.9 ± 7.6	8.6	68.0	105.7
NCH (cm)	15.8 ± 3.4	21.8	9.3	27.7
NC _{25%} (cm)	86.4 ± 7.3	8.4	71.3	105.0
NC _{50%} (cm)	111.9 ± 10.1	9.0	85.0	139.7
NC _{75%} (cm)	126.3 ± 8.6	6.8	104.3	147.0
Body ratios				
GC : HW	1.21 ± 0.05	4.29	1.09	1.39
WC : HW	1.25 ± 0.07	5.44	1.07	1.46
GC : BL	1.20 ± 0.04	3.75	1.11	1.37
WC : BL	1.24 ± 0.06	4.93	1.07	1.39
Neck ratios				
NC _{25%} : HW	0.55 ± 0.04	7.86	0.45	0.64
NC _{50%} : HW	0.71 ± 0.06	8.32	0.54	0.85
NC _{75%} : HW	0.80 ± 0.05	6.24	0.66	0.91
NC _{25%} : BL	0.54 ± 0.05	8.84	0.44	0.65
NC _{50%} : BL	0.70 ± 0.06	9.09	0.56	0.85
NC _{75%} : BL	0.79 ± 0.06	7.05	0.68	0.94
NC _{25%} : NL	0.98 ± 0.08	8.71	0.71	1.24
NC _{50%} : NL	1.26 ± 0.11	8.40	0.96	1.59
NC _{75%} : NL	1.43 ± 0.10	7.08	1.15	1.74
NC _{25%} : NCH	5.64 ± 0.96	17.08	3.72	8.43
NC _{50%} : NCH	7.28 ± 1.15	15.80	4.95	10.20
NC _{75%} : NCH	8.26 ± 1.43	17.31	5.31	11.93

HW = height at the withers; BL = body length; GC = girth circumference; WC = waist circumference; NL = neck length; NCH = neck crest height; NC_{25%} = neck circumference at 25% of neck length; NC_{50%} = neck circumference at 50% of neck length; NC_{75%} = neck circumference at 75% of neck length.

was observed in 97.5% of obese and 59.6% of non-obese horses (Table 2).

Statistical description of the morphometric measurements and calculated ratios are shown in Table 3. In general, all variables were homogenous, except the variables related with the NCH, which showed large variability evidenced by high CVs.

Influence of demographic variables on morphometric measurements

The effect of sex and age on morphometric parameters are shown in Table 4. The two circumferences over the trunk were greater in mares than in stallions ($P < 0.001$), while the stallions presented higher values in all measurements evaluated in the neck area ($P < 0.001$). The age affected BL, GC, WC, NCH and NC_{25%} showing in adult horses higher values ($P < 0.050$).

Morphometric ratios using the GC and the WC adjusted for body size (divided by HW and BL) showed higher values in the case of mares ($P < 0.001$). Similar results were observed

when the NCs were adjusted for the NCH. On the contrary, stallions significantly surpassed mares in all neck ratios adjusted for the HW and the BL ($P < 0.001$). Only two ratios (WC : HW and NC_{75%} : NCH) demonstrated differences according to age. Despite all neck measurements having been affected by the gender, when the three NCs were adjusted for the NL, the resulting ratios were independent of both demographic variables considered in the analysis (Table 4).

Influence of adiposity on morphometric measurements

Among all absolute measurements, only the GC and the NC_{50%} were not influenced by overall or regional adiposity (Table 5). The WC was significantly affected only by the obesity status, presenting greater values in those horses with $BCS \geq 7$ ($P = 0.021$). In the case of HW, NC_{25%} and NC_{75%}, these measurements were significantly higher in horses with cresty neck.

Only the ratios including GC and WC adjusted for the HW showed significant differences according to the obesity status, being greater than the values in the obese horse group. Exclusively the ratio NC_{75%} : BL was significantly associated with cresty neck condition. The rest of the calculated ratios were similar between both adiposity groups (Table 5).

Furthermore, several interactions between sex and adiposity groups could be observed in the multivariate analysis (Tables 6 and 7). Thereby, despite GC, NCs, GC : BL, NC_{25%} : HW and NCs : BL globally having shown significant differences by sex (Table 4), but not by adiposity status (Table 5), significantly greater values for the above parameters were observed in obese mares respect to the other three categories (Table 6).

Likewise, in the case of neck morphometry, stallions with cresty neck presented significantly higher values for NCs, NC_{25%} : HW, NCs : BL and NC_{25%} : NL (Table 7) while globally these measurements (except the last one) were significantly greater in stallions (Table 4) and only NC_{25%} and NC_{75%} were higher in horses with cresty neck condition (Table 5).

Discussion

In horses, the application of body measurements has been traditionally used to define their characteristics and general body conformation (Sadek *et al.*, 2006). Although previous studies have shown a clear distinction in morphometric measurements depending on the breed evaluated (Martinson *et al.*, 2014; Catalano *et al.*, 2016) as well as the influence of gender and age in the values obtained (Pinto *et al.*, 2008; Purzyc *et al.*, 2011), these differences mainly refer to simple measurements of conformational traits. However, in the clinical context, morphometrics have recently emerged as alternative estimators of obesity due to its relationship with scoring systems, body fat percentage and biochemical variables relevant to assess different aspects of metabolism (Henneke *et al.*, 1983; Carter *et al.*, 2009a). In this context, Frank *et al.* (2006) suggested that mean NC could be useful to identify horses at risk of insulin resistance. Afterwards, in a study conducted in horses and ponies some morphometric

Table 4 Statistical association of morphometric measurements (cm) and ratios with demographic variables

Variable	Sex			Age groups		
	Stallions	Mares	P-value	2 to 5 years	6 to 15 years	P-value
<i>n</i>	78	76		77	77	
Absolute measures						
HW (cm)	155.9 ± 1.54	155.8 ± 1.26	0.973	155.3 ± 1.47	156.4 ± 1.30	0.330
BL (cm)	158.7 ± 1.87	161.5 ± 1.53	0.051	158.4 ± 1.79	161.8 ± 1.58	0.008
GC (cm)	186.5 ± 2.31	196.2 ± 1.89	<0.001 ^a	189.4 ± 2.21	193.3 ± 1.96	0.013
WC (cm)	192.4 ± 3.00	206.4 ± 2.45	<0.001 ^a	196.9 ± 2.87	201.9 ± 2.54	0.015
NL (cm)	89.1 ± 2.38	84.1 ± 1.94	0.004	86.4 ± 2.27	86.8 ± 2.01	0.815
NCH (cm)	16.5 ± 0.98	13.6 ± 0.79	<0.001 ^a	14.3 ± 0.92	15.8 ± 0.83	0.030
NC _{25%} (cm)	87.9 ± 1.49	81.2 ± 1.22	<0.001 ^a	83.4 ± 1.42	85.7 ± 1.26	0.025
NC _{50%} (cm)	114.9 ± 2.22	105.0 ± 1.81	<0.001 ^a	108.5 ± 2.12	111.4 ± 1.88	0.060
NC _{75%} (cm)	128.0 ± 1.94	120.7 ± 1.58	<0.001 ^a	124.0 ± 1.85	124.7 ± 1.64	0.565
Body ratios						
GC : HW	1.20 ± 0.013	1.26 ± 0.011	<0.001 ^a	1.22 ± 0.013	1.24 ± 0.011	0.061
WC : HW	1.24 ± 0.017	1.32 ± 0.014	<0.001 ^a	1.27 ± 0.016	1.29 ± 0.014	0.034
GC : BL	1.17 ± 0.013	1.21 ± 0.011	<0.001 ^a	1.19 ± 0.013	1.19 ± 0.011	0.916
WC : BL	1.21 ± 0.018	1.28 ± 0.014	<0.001 ^a	1.24 ± 0.017	1.25 ± 0.015	0.613
Neck ratios						
NC _{25%} : HW	0.56 ± 0.010	0.52 ± 0.008	<0.001 ^a	0.54 ± 0.010	0.55 ± 0.009	0.107
NC _{50%} : HW	0.74 ± 0.015	0.67 ± 0.012	<0.001 ^a	0.70 ± 0.014	0.71 ± 0.012	0.202
NC _{75%} : HW	0.82 ± 0.013	0.77 ± 0.011	<0.001 ^a	0.80 ± 0.012	0.80 ± 0.011	0.883
NC _{25%} : BL	0.55 ± 0.010	0.50 ± 0.008	<0.001 ^a	0.53 ± 0.010	0.53 ± 0.009	0.576
NC _{50%} : BL	0.72 ± 0.014	0.65 ± 0.012	<0.001 ^a	0.69 ± 0.014	0.69 ± 0.012	0.700
NC _{75%} : BL	0.81 ± 0.013	0.75 ± 0.010	<0.001 ^a	0.78 ± 0.012	0.77 ± 0.011	0.170
NC _{25%} : NL	0.99 ± 0.027	0.97 ± 0.022	0.420	0.97 ± 0.026	0.99 ± 0.023	0.236
NC _{50%} : NL	1.29 ± 0.033	1.26 ± 0.027	0.130	1.26 ± 0.032	1.29 ± 0.028	0.220
NC _{75%} : NL	1.44 ± 0.033	1.44 ± 0.027	0.967	1.44 ± 0.032	1.44 ± 0.028	0.894
NC _{25%} : NCH	5.51 ± 0.312	6.12 ± 0.300	0.010	5.99 ± 0.293	5.64 ± 0.264	0.100
NC _{50%} : NCH	7.19 ± 0.373	7.87 ± 0.300	0.015	7.75 ± 0.351	7.31 ± 0.316	0.084
NC _{75%} : NCH	8.00 ± 0.444	9.05 ± 0.358	0.002 ^a	8.86 ± 0.418	8.19 ± 0.376	0.028

HW = height at the withers; BL = body length; GC = girth circumference; WC = waist circumference; NL = neck length; NCH = neck crest height; NC_{25%} = neck circumference at 25% of neck length; NC_{50%} = neck circumference at 50% of neck length; NC_{75%} = neck circumference at 75% of neck length.

^aPower value greater than 80%

ratios related to neck and overall adiposity were correlated with basal insulin, glucose, leptin and triglyceride concentrations (Carter *et al.*, 2009a).

Nowadays, little information is available about the use of morphometrics to characterize the obesity in equids. To the authors' knowledge, this is the first study where the effects of demographic variables and regional and general obesity scores were taken into account, to establish morphometric ratios related to the body condition specifically in Andalusian horses.

Notwithstanding, Andalusian horses are considered a light breed, the average of BCS observed in the present study is similar to those described for draft breeds (6.3 ± 0.9) (Catalano *et al.*, 2016) and exceed the values reported in previous studies related to light breeds (5.38 ± 0.72) (Wagner and Tyler, 2011). Likewise, the proportion of obese horses was also higher than values reported in other breeds where the same cut-off value was used to determine this condition (Harker *et al.*, 2011; Jensen *et al.*, 2016). Similarly, the proportion of cresty neck horses was also higher than in

other breeds (Giles *et al.*, 2015). In relation with these findings and as expected, mean values of several morphometrics such as GC (Sadek *et al.*, 2006; Čoudková *et al.*, 2016), WC (McGowan *et al.*, 2013), NC_{50%} (Bailey *et al.*, 2008; Martinson *et al.*, 2014; Catalano *et al.*, 2016) and NCH (Wray *et al.*, 2013) were greater compared with previously published data in light horse breeds and ponies. Furthermore, the CV of the measurements made of the trunk and neck area proved to be quite homogeneous, although less than those described in Arabian horses (Sadek *et al.*, 2006). The differences between Andalusian and other light breed horses may be related to the morphologic standard sought by breeders or, by the idiosyncrasy of the breed.

Influence of demographic variables on morphometric measurements

The Andalusian horse is described as a breed with a significant sexual dimorphism (Stud breeding book of Andalusians, 2002), this term refers to differences in dimensions (absolute measurements) and proportions (ratios) of the body

Table 5 Statistical association of morphometric measurements (cm) and ratios with obesity status and cresty neck condition

Variable	Obesity status			Cresty neck condition		
	BCS < 7	BCS ≥ 7	P-value	CNS < 3	CNS ≥ 3	P-value
<i>n</i>	114	40		47	107	
Absolute measures						
HW (cm)	156.8 ± 0.53	154.9 ± 2.47	0.462	152.8 ± 2.51	158.9 ± 0.49	0.018
BL (cm)	158.9 ± 0.65	161.3 ± 3.00	0.425	159.7 ± 3.05	160.5 ± 0.59	0.816
GC (cm)	188.4 ± 0.80	194.3 ± 3.71	0.117	188.3 ± 3.76	194.33 ± 0.73	0.116
WC (cm)	193.8 ± 1.03	205.1 ± 4.81	0.021	198.9 ± 4.88	199.9 ± 0.95	0.835
NL (cm)	88.9 ± 0.82	84.3 ± 3.81	0.229	83.9 ± 3.87	89.3 ± 0.75	0.172
NCH (cm)	15.6 ± 0.35	14.5 ± 1.55	0.499	13.9 ± 1.58	16.2 ± 0.31	0.143
NC _{25%} (cm)	84.5 ± 0.51	84.6 ± 2.38	0.990	81.9 ± 2.42	87.2 ± 0.47	0.033
NC _{50%} (cm)	109.8 ± 0.77	110.1 ± 3.56	0.927	106.6 ± 3.61	113.4 ± 0.70	0.063
NC _{75%} (cm)	124.0 ± 0.67	124.7 ± 3.11	0.805	120.7 ± 3.15	128.0 ± 0.61	0.023
Body ratios						
GC : HW	1.20 ± 0.005	1.25 ± 0.022	0.015	1.23 ± 0.022	1.22 ± 0.004	0.637
WC : HW	1.24 ± 0.006	1.33 ± 0.027	0.001 ^a	1.30 ± 0.027	1.26 ± 0.005	0.109
GC : BL	1.19 ± 0.005	1.20 ± 0.021	0.398	1.18 ± 0.021	1.21 ± 0.004	0.121
WC : BL	1.22 ± 0.006	1.27 ± 0.028	0.081	1.24 ± 0.029	1.25 ± 0.006	0.846
Neck ratios						
NC _{25%} : HW	0.54 ± 0.003	0.55 ± 0.016	0.662	0.54 ± 0.016	0.55 ± 0.003	0.477
NC _{50%} : HW	0.70 ± 0.005	0.71 ± 0.023	0.640	0.70 ± 0.024	0.71 ± 0.005	0.531
NC _{75%} : HW	0.79 ± 0.005	0.80 ± 0.021	0.496	0.79 ± 0.021	0.81 ± 0.004	0.480
NC _{25%} : BL	0.53 ± 0.004	0.52 ± 0.016	0.617	0.51 ± 0.017	0.54 ± 0.003	0.054
NC _{50%} : BL	0.69 ± 0.005	0.68 ± 0.023	0.712	0.67 ± 0.023	0.71 ± 0.004	0.074
NC _{75%} : BL	0.78 ± 0.004	0.77 ± 0.020	0.746	0.76 ± 0.021	0.80 ± 0.004	0.039
NC _{25%} : NL	0.95 ± 0.009	1.01 ± 0.043	0.219	0.98 ± 0.044	0.98 ± 0.009	0.931
NC _{50%} : NL	1.24 ± 0.012	1.31 ± 0.054	0.189	1.28 ± 0.054	1.27 ± 0.011	0.958
NC _{75%} : NL	1.40 ± 0.011	1.48 ± 0.053	0.121	1.45 ± 0.054	1.44 ± 0.011	0.889
NC _{25%} : NCH	5.61 ± 0.111	6.03 ± 0.492	0.398	6.11 ± 0.502	5.53 ± 0.097	0.253
NC _{50%} : NCH	7.26 ± 0.133	7.80 ± 0.590	0.364	7.89 ± 0.601	7.17 ± 0.116	0.238
NC _{75%} : NCH	8.23 ± 0.159	8.83 ± 0.702	0.396	8.94 ± 0.715	8.12 ± 0.138	0.259

BCS = body condition score; CNS = cresty neck score; HW = height at the withers; BL = body length; GC = girth circumference; WC = waist circumference; NL = neck length; NCH = neck crest height; NC_{25%} = neck circumference at 25% of neck length; NC_{50%} = neck circumference at 50% of neck length; NC_{75%} = neck circumference at 75% of neck length.

^aPower value greater than 80%.

Table 6 Statistical association of morphometric measurements (cm) and ratios with interactions between sex and obesity status

Variable	Stallions		Mares		P-value
	BCS < 7	BCS ≥ 7	BCS < 7	BCS ≥ 7	
<i>n</i>	57	21	57	19	
Absolute measures					
GC (cm)	185.2 ± 1.27	187.8 ± 4.21	191.6 ± 0.95	200.7 ± 3.66	0.027
NC _{25%} (cm)	89.1 ± 0.82	86.7 ± 2.71	80.0 ± 0.61	82.5 ± 2.36	0.008
NC _{50%} (cm)	116.2 ± 1.22	113.6 ± 4.04	103.4 ± 0.92	106.6 ± 3.52	0.040
NC _{75%} (cm)	129.0 ± 1.06	127.1 ± 3.53	118.9 ± 0.80	122.4 ± 3.07	0.027
Body ratios					
GC : BL	1.18 ± 0.007	1.17 ± 0.024	1.20 ± 0.005	1.23 ± 0.021	0.013
Neck ratios					
NC _{25%} : HW	0.57 ± 0.006	0.56 ± 0.018	0.51 ± 0.004	0.53 ± 0.016	0.029
NC _{25%} : BL	0.57 ± 0.006	0.54 ± 0.019	0.50 ± 0.004	0.51 ± 0.016	0.007
NC _{50%} : BL	0.74 ± 0.008	0.71 ± 0.026	0.65 ± 0.006	0.66 ± 0.022	0.023
NC _{75%} : BL	0.82 ± 0.007	0.79 ± 0.023	0.74 ± 0.005	0.75 ± 0.020	0.019

BCS = body condition score; GC = girth circumference; NC_{25%} = neck circumference at 25% of neck length; NC_{50%} = neck circumference at 50% of neck length; NC_{75%} = neck circumference at 75% of neck length; BL = body length; HW = height at the withers.

Table 7 Statistical association of morphometric measurements (cm) and ratios with interactions between sex and cresty neck condition

Variable	Stallions		Mares		P-value
	CNS < 3	CNS ≥ 3	CNS < 3	CNS ≥ 3	
<i>n</i>	9	69	38	38	
Absolute measures					
NC _{25%} (cm)	83.5 ± 2.86	92.3 ± 0.58	80.4 ± 2.30	82.1 ± 0.73	0.001
NC _{50%} (cm)	110.0 ± 4.27	119.9 ± 0.87	103.2 ± 3.43	106.9 ± 1.09	0.044
NC _{75%} (cm)	122.9 ± 3.73	133.2 ± 0.76	118.5 ± 2.99	122.8 ± 0.95	0.025
Neck ratios					
NC _{25%} : HW	0.55 ± 0.019	0.58 ± 0.004	0.52 ± 0.016	0.52 ± 0.005	0.007
NC _{25%} : BL	0.53 ± 0.020	0.58 ± 0.004	0.50 ± 0.016	0.51 ± 0.005	0.001 ^a
NC _{50%} : BL	0.69 ± 0.027	0.75 ± 0.006	0.64 ± 0.022	0.66 ± 0.007	0.030
NC _{75%} : BL	0.77 ± 0.024	0.84 ± 0.005	0.74 ± 0.020	0.76 ± 0.006	0.017
NC _{25%} : NL	0.97 ± 0.052	1.01 ± 0.011	1.00 ± 0.042	0.95 ± 0.013	0.037

CNS = cresty neck score; NC_{25%} = neck circumference at 25% of neck length; NC_{50%} = neck circumference at 50% of neck length; NC_{75%} = neck circumference at 75% of neck length; BL = body length; NL = neck length.

^aPower value greater than 80%.

between males and females (Glucksmann, 1974). This aspect has been corroborated in an objective way, since most of the absolute morphometric measurements evaluated in this study were significantly different between both sexes. The higher values observed in neck morphometrics, in the case of males compared with females, may be explained due to the fact that the neck crest area contains great amounts of connective tissue within the adipose tissue (Dugdale *et al.*, 2011a). This may contribute to a possible functional and sexually dichotomous role (Dugdale *et al.*, 2011a). In regard to the measurements taken of the trunk, the greater circumferences values obtained in females compared with males, are similar to results reported in other horse breeds (Rastija *et al.*, 2004; Sadek *et al.*, 2006). In addition, owing to the higher values obtained by mares in GC and WC measurements; resulting mean values of all body ratios were higher and significantly different between both genders. This indicates that these ratios in a male cannot be compared with the same measurement in a female, as the same value would not imply similar adiposity. At this point, it is interesting to note that, despite the proportion of obese animals assessed by BCS being similar between males and females; when morphometric measurements were used to evaluate the population, gender-related differences in the neck and body condition were highlighted. The differences according to age could be attributed to the change in conformation, muscular development and increase in the fat deposit that takes place in horses as they reach adulthood (Martin-Rosset *et al.*, 2008; Čoudková *et al.*, 2016).

Influence of adiposity on morphometric measurements

In contrast with those studies showing that GC is affected by adiposity (Thatcher *et al.*, 2012), in our case GC was not influenced by adiposity status. In humans, the measurement of WC is a better indicator of abdominal fat accumulation than the body mass index (Lee *et al.*, 2006). In the case of equids, it has been shown that considerable losses in the

body mass corresponded to disproportionately small or even absent changes on BCS while morphometrics like the WC were more accurate indexes for assessing and monitoring changes in adiposity (Dugdale *et al.*, 2010, 2011a and 2011b). The fact that the WC was in relation with differences in body condition in Andalusians reinforces the sensitivity of this measurement.

Absolute morphometric measurements alone may be uninformative for assessing relative adiposity, thus morphometric ratios that have been shown to be useful for the evaluation of body condition in equids, include the use of a trunk or NC in relation to HW and BL. These ratios represent more than one physical dimension and therefore are more sensitive to compare fat deposits than any individual physical measure (Henneke *et al.*, 1983). Recently, the GC : HW ratio has been suggested as the most appropriate objective measure to assess overall adiposity in horses (Carter *et al.*, 2009a). In agreement with this observation, in our case the GC : HW but also WC : HW were the only ratios related to general obesity independently of the neck adiposity. Otherwise, different cut-off values for this ratio have been proposed to estimate an overweight status depending on the horse breed evaluated (Carter *et al.*, 2009a; Jensen *et al.*, 2016). The mean value of GC : HW ratio in obese Andalusians was lower than the values described in the literature for overweight (1.27) and obese (1.29) Icelandic horses (Jensen *et al.*, 2016). These discrepancies could be probably attributed to the morphology of the horses included and, stand out the need to re-adjust the cut-off values according to each breed specific criteria.

Several morphometric measurements have been described specifically for the assessment of fat deposition along the neck (Frank *et al.*, 2006; Bailey *et al.*, 2008; Martin-Gimenez *et al.*, 2016). Among them points out the use of the NCs due to its associations with the CNS (Carter *et al.*, 2009a). Similarly, in this case, the high percentage of cresty neck horses observed during the study, and that characterize Andalusian

purebred horses, corresponded with the large NCs values registered. However, we cannot make the assumption that an increase in their diameter is due to an increase in fat deposition as CNS can only assess the external appearance of the neck and muscle development and conformation may be influencing the scoring process. Moreover, unlike the observations obtained in Icelandic horses where NC_{50%} increased when the BCS increased (Jensen *et al.*, 2016), in our study all neck measurements were similar between obese and non-obese horses.

On the other hand, it has been proposed that the NC_{50%}:HW ratio could be used as an alternative adiposity measurement when a horse obtains a value ≥ 0.63 (equivalent to a CNS ≥ 3) (Carter *et al.*, 2009a). Nonetheless, our global mean values as well as the values registered in the obese and cresty neck groups exceeded not only the threshold mentioned above, but also the cut-off value (>0.71) associated with a greater predisposition of developing an endocrinopathic laminitis (Carter *et al.*, 2009b). In relation with this, in our study only one mare with value >0.71 for the NC_{50%}:HW ratio ($=0.75$) had an owner-reported history of laminitis.

Even when the three NCs were put into context by relating them to all neck or body size measures, the resulting ratios showed their stability. This along with the fact that more than a half of the horses classified as non-obese exhibited a cresty neck appearance suggest that at least in this breed, great neck morphometrics are more related to a phenotypic feature and cannot be used to discriminate among obese and non-obese horses.

Finally, similarly to what happens in humans, where the morphometric criteria to define a man as obese varies in comparison with a woman (Lin *et al.*, 2002), the interactions observed in our study verify that many morphometric values significantly differ depending on the body condition of males or females.

In our attempt to find out suitable morphometric parameters for assessment of overall and regional adiposity, a series of absolute measurements and ratios were studied here, showing that many breed related morphological characteristics were influencing this evaluation. These results emphasize the necessity to set specific reference values according to the gender and adiposity condition in different breeds.

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