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Pinch size can affect the skinfold thickness measurement and interfere in the estimation and classification of body adiposity

O tamanho da pinça pode afetar a medida de espessura das dobras cutâneas e interferir na estimativa e classificação da adiposidade corporal

Joaquim Huaina Cintra-Andrade[1]
https ://orcid.org/0000-0001-8614-2939
Filipe Oliveira de Brito[2]
https ://orcid.org/0000-0002-0234-9455
Maria Izabel Freire-Correia[2]
https ://orcid.org/0000-0001-5130-6796
Roberto Fernandes da Costa[3]
https ://orcid.org/0000-0002-8789-1744
Wagner Luis Ripka[4]
https ://orcid.org/0000-0002-6191-1188

1. Ceará State University. Fortaleza, CE. Brazil.
2. University of Fortaleza. Fortaleza, CE. Brazil.
3. Federal University of Rio Grande do Norte. Natal, RN. Brazil.
4. Federal Technological University - Paraná. Curitiba, PR. Brazil.

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Corresponding author
Joaquim Huaina Cintra-Andrade
Centro de Ciências da Saúde,
Universidade Estadual do Ceará - UECE
Av. Dr. Silas Munguba, 1700 – Itaperi - 60714-903. Fortaleza/CE, Brasil.
E-mail: joaquimcintra@hotmail.com

Abstract - The aim of this study was to verify the effect of pinch size on skinfold thickness measurement and the consequent interference in the estimation and classification of body adiposity components. Cross-sectional and quantitative study carried out with a sample of 29 subjects recruited from a university in the city of Fortaleza, Ceará, Brazil. Four measurement steps were performed at each site of the eight chosen skinfolds. The first step was performed with a subjective-landmark and the three subsequent steps with fixed-landmarks defined with an expanding secondary line at 2 cm intervals. Body adiposity components were determined from the skinfold thickness measured at each landmark. Repeated measures ANOVA and Bland-Altman agreement analysis were applied. The subjective-landmark was chosen as the dependent variable. The 6 cm-landmark showed similarity and statistical agreement with the subjective-landmark for all skinfolds except the thigh, and with the sums of five and eight skinfolds. All fixed-landmarks showed agreement below the cut-off point for the percentile classification of subcutaneous adiposity and normative relative body fat. Variation in pinch size is an important source of TEM that can affect the reproducibility of skinfold thickness measurements and interfere in the estimation and classification of the molecular and tissue component of body adiposity.

Key words: Anthropometry; Skinfold thickness; Adiposity; Body composition.

Resumo – *O objetivo deste estudo foi verificar o efeito do tamanho da pinça na medida de espessura das dobras cutâneas e a consequente interferência na estimativa e classificação dos componentes da adiposidade corporal. Estudo transversal e quantitativo realizado com amostra de 29 sujeitos recrutados em uma universidade da cidade de Fortaleza, Ceará, Brasil. Quatro etapas de medição foram realizadas em cada sítio das oito dobras cutâneas escolhidas. A primeira etapa foi realizada com um marco subjetivo e as três etapas subsequentes com marcos fixos definidos com uma linha secundária expansiva em intervalos de 2 cm. Os componentes da adiposidade corporal foram determinados a partir da espessura de dobras cutâneas mensuradas em cada marco. ANOVA de medidas repetidas e análise de concordância de Bland-Altman foram aplicadas. O marco subjetivo foi escolhido como variável dependente. O marco de 6 cm apresentou semelhança e concordância estatística com o marco subjetivo para todas as dobras cutâneas, exceto a coxa, e com as somas de cinco e oito espessuras de dobras cutâneas. Todos os marcos fixos mostraram concordância abaixo do ponto de corte para a classificação percentílica de adiposidade subcutânea e gordura corporal relativa normativa. A variação no tamanho da pinça é uma importante fonte de ETM que pode afetar a reprodutibilidade de medida de espessura das dobras cutâneas e interferir na confiabilidade da estimativa e classificação do componente molecular e tecidual da adiposidade corporal.*

Palavras-chave: *Antropometria; Espessura da dobra cutânea; Adiposidade; Composição corporal.*

INTRODUCTION

Surface anthropometry is a satisfactorily valid doubly-indirect method to describe and quantify human body composition in different field settings[1]. Skinfold thickness is the main measurable property to estimate the tissue and/or molecular component of body adiposity. However, some biological limitations are attributed to skinfolds. Skin thickness and dynamic and static compressibility of subcutaneous adipose tissue differ considerably between sites and subjects[3]. Thus, the reproducibility and reliability of skinfold thickness is dependent on the anthropometrist's accuracy and adherence to the measurement technique[1].

The depth of application of skinfold caliper contact jaws and, more specifically, site location are well-documented sources of measurement error[4,5]. An important international association specializing in anthropometry emphasizes that, regardless of the degree of technical skill of the anthropometrist, all skinfold sites must be pre-identified and accurately marked[6]. A site is the anatomical location for skinfold measurement, where a landmark is performed with two intersecting lines. The primary line corresponds to the direction of the vertical, oblique, or horizontal anatomical axis and the secondary line corresponds to the perpendicular position of the index and thumb fingers in a pinch shape[7].

The distance between the fingers is proportional to the size of the pinch needed to form a skinfold, however, it is described with divergence in the reference literature. Brozek and Keys[8] and Harrison et al.[9] while recognizing the importance of technical-palpatory subjectivity, suggest about 8 cm as the standard distance for pinching a skinfold. Ross and Marfell-Jones[10] and Esparza-Ros et al.[6] only describe that the distance between the fingers is strictly subjective and that it be sufficient to ensure the formation of a parallel layer of skin and subcutaneous adipose tissue. The effect of pinch size on the measurement of skinfold thickness hasn't been keenly investigated. Although there is no experimental evidence, it is hypothesized that the way that skinfold thickness is pinched may increase the degree of variability in the measurement[1,11]. Thus, the present study aimed to verify the effect of pinch size on skinfold thickness measurement and the consequent interference in the estimation and classification of body adiposity components.

METHODS

Participants

Cross-sectional and quantitative study carried out in the last quarter of 2021 at a university in the city of Fortaleza, Ceará, Brazil. The non-probabilistic convenience sample consisted of 29 subjects of both sexes randomly recruited. Subjects aged 20 to 35 years and self-reported as healthy were chosen. Subjects who had undergone liposuction surgery and/or abdominoplasty were excluded. In addition, subjects were excluded if during the collection session any skinfold was biologically impossible to measure. The subjects' participation was voluntary and the informed consent form was signed. The study followed the Brazil's National Health Council's research guidelines involving human experimentation. Approval was obtained by the Ethics and Research Committee of Platform Brazil under the University of Fortaleza, with number: CAAE - 89306918.9.0000.5052

Procedures

An anthropometrist accredited at level 3 by the International Society for the Advancement of Kinanthropometry (ISAK) was selected to perform the anthropometric measurements in a private room at a temperature of 24°C, employing the *International Standards for Anthropometric Assessment*[6]. Body mass was measured using a digital scale (Toledo®, 2098PP, Brazil) and height using a stadiometer (Sanny®, ES2030, BR). The triceps, subscapular, biceps, iliac crest, supraspinale, abdominal, thigh, and calf skinfold thicknesses were measured using a Harpenden® skinfold caliper (Baty International®, UK) with downward static pressure of 8.25 g/mm^2. The caliper dial indicator was pre-calibrated using a 10 mm steel gauge block (Digimess®, 150.499-81, BR). The site and landmark of the skinfolds were identified and marked using a flexible steel anthropometric tape (Cescorf®, BR), an anthropometric box (Anthropos®, BR) and a dermographic pen (Viscot®, USA).

For positioning of the caliper jaws, a short guideline[12] was added 1 cm away from the intersection and in the direction of the anatomical axis[6]. Four measurement steps were performed at each site of the eight skinfolds. The first step was performed with a subjective-landmark and the three subsequent steps with fixed-landmarks defined with a secondary line centered at the intersection of the site and expanding outwards at 2 cm intervals (Figure 1). The effect of skinfold compressibility[3] was minimized with a 10-minute interval between measurement steps. In the first step, the distance between the fingers was defined subjectively as described in Esparza-Ros et al.[6]. The chosen pinch size was marked with two dots immediately above the perpendicular/secondary line to the anatomical axis. In the second step, a fixed distance of 4 cm was marked. In the third step, a fixed distance of 6 cm was marked. Finally, in the fourth step, a fixed distance of 8 cm was marked.

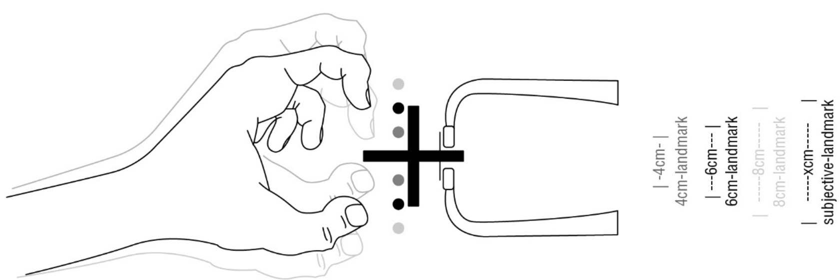

Figure 1. Illustrative figure on the differences in the size of the landmark in a skinfold site related to the positioning of 4 cm, 6 cm, 8 cm and subjective landmark.

A duplicate was performed at each landmark of the eight skinfolds. The mean value was used for statistical analyses. In the event of an error >5%, a triplicate was performed and, consequently, the intermediate value was used. The intra-evaluator relative Technical Error of Measurement (TEM) was calculated[13] and presented in Table 1. Components of body adiposity were determined from the skinfold thickness measured at each landmark. *Molecular component*: body density was determined from mathematical models nºM7 and nºF9 proposed by Petroski[14] for males and females, respectively. The value was converted to relative body fat[15] and classified[16]. *Tissue component*: subcutaneous adiposity was determined, in absolute values, from the sum of five skinfolds (triceps, subscapular, iliac crest, abdominal and thigh). Percentile curves were applied for classification[17].

Statistical analysis

Descriptive statistics were applied. Normality of the data was analyzed using the Shapiro-Wilk test. Differences between skinfold thicknesses obtained at each landmark were analyzed with analysis of variance (ANOVA) for repeated measures. Skinfold measurements that did not follow the normal distribution were compared using the Friedman test and presented as medians and interquartile range. The subjective-landmark was defined as a dependent variable and compared with the fixed-landmarks using the Bland-Altman technique. This statistical procedure quantifies measures of agreement by bias and limits of agreement (LOA). The existence of proportional bias was analyzed using the Ordinary Least Square (OLS) regression model, using as independent variable the mean value of the value measured by the compared techniques and as dependent variable the value of the difference between the compared measures. The fixed bias was established with the one-sample t test for the values of differences between measurements[18].

The difference in the number of subjects in each classification of body adiposity components, either by percentile or relative value, was verified using the Chi-Square test. The agreement between the classification parameters was analyzed using the Kappa coefficient. Value $\kappa \geq 0.8$ was considered. The significance level was set at $p<0.05$. IBM SPSS Statistics, version 26.0 (IBM Corp., Armonk, NY, USA) was used for all analysis.

RESULTS

The sample consisted of 29 subjects (51.7% women) with a mean age of 26.48±3.48 years. Each subject was evaluated on all variables. The mean relative body fat and the sum of five skinfolds were 19.51±5.65% and 79.80±25.48 mm, respectively. The results are classified between the 25th and 50th percentiles, characterizing the sample as eutrophic[16,17].

The comparative analysis between the landmarks is shown in Table 1. The 4 cm-landmark presented similarity with the subjective-landmark for the triceps, supraspinale and calf skinfolds, as well as with the 6 cm-landmark for the triceps, supraspinale, thigh, and calf skinfolds, and with the 8 cm-landmark for the subscapular skinfold. The 6 cm-landmark showed similarity with the subjective-landmark for the skinfolds and the sum of the thickness of five and eight skinfolds. The 8 cm-landmark showed similarity with the subjective-landmark for supraspinal and abdominal skinfolds. Furthermore, a significant difference was observed between the landmarks for the relative body fat component, except between the 6 cm-landmark and the 8 cm-landmark (Table 1).

The Bland-Altman agreement analysis between the subjective-landmark and the fixed-landmarks is shown in Table 2. No fixed-landmark presented satisfactory LOA for all skinfold thicknesses. The 4 cm-landmark agreed with the subjective-landmark for triceps and calf skinfolds. The 6 cm-landmark agreed with the subjective-landmark for all skinfolds, except the thigh. It was also the only fixed-landmark that showed agreement with the sums of skinfolds. The 8 cm-landmark agreed with the subjective-landmark for supraspinale, abdominal and thigh skinfolds. All fixed-landmarks showed significant fixed bias for the estimation of relative body fat (Figure 2). The absolute and relative frequency of the classification of body adiposity components between the subjective-landmark and the fixed-landmarks is presented in Table 3. There was

Table 1. Descriptive characteristics of the sample and comparison between landmarks.

Variables	Mean ± SD (TEM%)				p-value					
	A	B	C	D	A vs B	A vs C	A vs D	B vs C	B vs D	C vs D
	Subjective landmark	4 cm landmark	6 cm landmark	8 cm landmark						
Triceps skinfold (mm)	13.58 ± 5.02 (1.52%)	13.45 ± 5.08 (1.65%)	13.73 ± 5.19 (1.48%)	14.33 ± 5.43 (1.90%)	1.000	1.000	**0.003**	0.954	**0.025**	**0.002**
Subscapular skinfold (mm)	12.24 ± 3.54 (2.18%)	11.84 ± 3.33 (1.99%)	12.21 ± 3.5 (2.93%)	12.8 ± 3.89 (1.92%)	**0.002**	1.000	**0.009**	**0.001**	0.191	**0.002**
Biceps skinfold (mm)	6.20 ± 5.00 (2.66%)	6.00 ± 4.20 (2.93%)	6.30 ± 4.90 (2.83%)	7.40 ± 4.60 (3.94%)	**0.002**	0.089	**0.002**	**0.001**	**<0.001**	**0.001**
Iliac crest skinfold (mm)	16.51 ± 6.42 (2.07%)	15.84 ± 5.98 (1.21%)	16.72 ± 6.21 (1.42%)	17.66 ± 6.98 (2.40%)	**0.018**	1.000	**0.034**	**<0.001**	**0.001**	**0.040**
Supraspinale skinfold (mm)	9.80 ± 6.85 (2.59%)	10.20 ± 6.10 (2.30%)	9.40 ± 18.00 (2.19%)	10.30 ± 6.75 (2.19%)	0.054	0.415	0.092	0.139	**0.012**	**0.002**
Abdominal skinfold (mm)	19.18 ± 6.18 (1.32%)	17.27 ± 5.18 (2.92%)	19.09 ± 6.36 (1.04%)	20.25 ± 7.45 (0.94%)	**0.001**	1.000	1.000	**0.001**	**0.001**	**0.004**
Thigh skinfold (mm)	18.70 ± 9.95 (1.62%)	17.00 ± 8.35 (1.05%)	18.00 ± 9.60 (1.04%)	19.00 ± 9.05 (1.55%)	**0.003**	0.148	**0.021**	0.070	**<0.001**	**<0.001**
Calf skinfold (mm)	12.39 ± 4.87 (1.20%)	12.18 ± 4.77 (2.32%)	12.48 ± 4.9 (1.33%)	12.95 ± 5.2 (1.92%)	0.440	1.000	**0.019**	0.051	**0.006**	**0.049**
Sum of 5 skinfolds (mm)	79.79 ± 4.73 *	75.79 ± 4.33 *	79.15 ± 4.51 *	83.80 ± 4.96 *	**<0.001**	1.000	**0.008**	**0.001**	**<0.001**	**<0.001**
Sum of 8 skinfolds (mm)	109.78 ± 34.58 *	104.73 ± 31.46 *	109.28 ± 32.89 *	115.47 ± 36.46 *	**<0.001**	1.000	**<0.001**	**<0.001**	**<0.001**	**<0.001**
Relative body fat (%)	19.51 ± 5.65 *	16.01 ± 5.28 *	17.12 ± 5.75 *	17.11 ± 5.41 *	**<0.001**	**0.006**	**0.009**	**<0.001**	**<0.001**	1.000

Note. SD: Standard deviation; TEM: Technical error of measurement. *TEM not calculated.

Table 2. Bland-Altman agreement analysis between the subjective-landmark and the fixed-landmarks.

Variables	Regression				One sample t-test			
	β	LOA	p-value	Bias*	Mean difference (±SEM)	LOA	p-value	Bias**
Triceps skinfold								
4 cm-landmark	-0.055	-0.098; 0.074	0.778	No	0.13 ± 0.2	-0.28; 0.55	0.519	No
6 cm-landmark	-0.255	-0.083; 0.017	0.182	No	-0.15 ± 0.12	-0.4; 0.11	0.245	No
8 cm-landmark	-0.397	-0.149; -0.007	**0.033**	Yes	-0.75 ± 0.19	-1.14; -0.36	**<0.0001**	Yes
Subscapular skinfold								
4 cm-landmark	0.384	-1.015; 0.388	**0.04**	Yes	0.4 ± 0.1	0.2; 0.6	**<0.0001**	Yes
6 cm-landmark	0.065	-0.050; 0.070	0.739	No	0.03 ± 0.1	-0.17; 0.23	0.77	No
8 cm-landmark	-0.415	-0.178; -0.013	**0.025**	Yes	-0.56 ± 0.16	-0.88; -0.23	**0.001**	Yes
Biceps skinfold								
4 cm-landmark	0.273	-0.785; 0.628	0.153	No	0.39 ± 0.13	0.12; 0.66	**0.006**	Yes
6 cm-landmark	0.104	-0.089; 0.153	0.592	No	-0.17 ± 0.16	-0.5; 0.15	0.286	No
8 cm-landmark	-0.049	-0.192; 0.150	0.801	No	-0.69 ± 0.23	-1.16; -0.23	**0.005**	Yes
Iliac crest skinfold								
4 cm-landmark	0.393	0.005; 0.137	**0.035**	Yes	0.67 ± 0.21	0.25; 1.1	**0.003**	Yes
6 cm-landmark	0.224	-0.025; 0.094	0.243	No	-0.21 ± 0.18	-0.58; 0.16	0.247	No
8 cm-landmark	-0.273	-0.203; 0.033	0.151	No	-1.14 ± 0.38	-1.93; -0.36	**0.006**	Yes
Supraspinale skinfold								
4 cm-landmark	0.483	0.033; 0.197	**0.008**	Yes	0.45 ± 0.2	0.03; 0.87	**0.035**	Yes
6 cm-landmark	0.309	-0.012; 0.124	0.103	No	0.12 ± 0.16	-0.21; 0.44	0.475	No
8 cm-landmark	-0.045	-0.110; 0.087	0.818	No	-0.44 ± 0.23	-0.9; 0.02	0.061	No
Abdominal skinfold								
4 cm-landmark	0.433	0.033; 0.335	**0.019**	Yes	1.91 ± 0.44	1.01; 2.81	**<0.0001**	Yes
6 cm-landmark	-0.055	-0.239; 0.181	0.777	No	0.09 ± 0.6	-1.14; 1.32	0.882	No
8 cm-landmark	-0.32	-0.438; 0.034	0.09	No	-1.08 ± 0.77	-2.64; 0.49	0.171	No

Note. SEM: standard error of the mean; β: coefficient of the least ordinary squares regression converted to z score; LOA: Bland-Altman limits of agreement; *Proportional bias if β differs significantly from 0 ($p<0.05$); **Fixed bias if p-value from t-test <0.05 or 95.

Table 2. Continued...

Variables	Regression				One sample t-test			
	β	LOA	p-value	Bias*	Mean difference (±SEM)	LOA	p-value	Bias**
Thigh skinfold								
4 cm-landmark	0.472	0.030; 0.198	**0.01**	Yes	0.89 ± 0.37	0.13; 1.65	**0.023**	Yes
6 cm-landmark	0.469	0.039; 0.261	**0.01**	Yes	0.88 ± 0.48	-0.1; 1.86	0.077	No
8 cm-landmark	0.313	-0.012; 0.133	0.098	No	-0.48 ± 0.3	-1.1; 0.15	0.128	No
Calf skinfold								
4 cm-landmark	0.153	-0.030; 0.070	0.428	No	0.21 ± 0.11	-0.02; 0.45	0.073	No
6 cm-landmark	-0.072	-0.045; 0.031	0.71	No	-0.08 ± 0.09	-0.26; 0.09	0.348	No
8 cm-landmark	-0.359	-0.134; 0.002	0.056	No	-0.56 ± 0.17	-0.91; -0.2	**0.003**	Yes
Sum of 5 skinfolds								
4 cm-landmark	0.563	0.037; 0.140	**0.001**	Yes	4.0 ± 0.71	2.54; 5.46	**<0.0001**	Yes
6 cm-landmark	0.234	-0.030; 0.125	0.223	No	0.64 ± 0.93	- 1.27; 2.55	0.499	No
8 cm-landmark	-0.212	-0.140; 0.041	0.27	No	-4.00 ± 1.12	- 6.30; - 1.69	**0.001**	Yes
Sum of 8 skinfolds								
4 cm-landmark	0.675	0.054; 0.136	**<0.0001**	No	5.06 ± 0.86	3.3; 6.81	**<0.0001**	Yes
6 cm-landmark	0.332	-0.006; 0.107	0.079	No	0.5 ± 0.95	-1.44; 2.44	0.601	No
8 cm-landmark	-0.296	-0.122; 0.015	0.119	No	-5.69 ± 1.19	-8.12; -3.26	**<0.0001**	Yes
Relative fat (%)								
4 cm-landmark	0.104	-0.218; 0.374	0.592	No	3.49 ± 0.72	2.03; 4.96	**<0.0001**	Yes
6 cm-landmark	-0.03	-0.272; 0.233	0.877	No	2.39 ± 0.65	1.07; 3.71	**0.001**	Yes
8 cm-landmark	0.071	-0.226; 0.325	0.714	No	2.4 ± 0.68	1.01; 3.79	**0.001**	Yes

Note. SEM: standard error of the mean; β: coefficient of the least ordinary squares regression converted to z score; LOA: Bland-Altman limits of agreement; *Proportional bias if β differs significantly from 0 ($p<0.05$); **Fixed bias if p-value from t-test <0.05 or 95.

Table 3. Analysis of the classification agreement of body adiposity components.

Classification	A	B	C	D	A vs B			A vs C			A vs D		
	Subjective landmark	4 cm landmark	6 cm landmark	8 cm landmark	χ^2	κ	p-value	χ^2	κ	p-value	χ^2	κ	p-value
Subcutaneous adiposity													
P5	5 (17.2%)	6 (20.7%)	8 (27.6%)	6 (20.7%)	**<0.0001**	0.489	**<0.0001**	**<0.0001**	0.668	**<0.0001**	**<0.0001**	0.759	**<0.0001**
P10	4 (23.8%)	4 (13.8%)	4 (13.8%)	4 (13.8%)									
P25	11 (37.9%)	10 (34.5%)	10 (34.5%)	11 (37.9%)									
P50	9 (31%)	7 (24.1%)	7 (24.1%)	8 (27.6%)									
Relative body fat													
Very low	0	1 (3.4%)	0	0	0.622	0.075	0.552	0.496	0.016	0.907	0.496	0.016	0.907
Low	16 (55.2%)	22 (75.9%)	21 (72.4%)	21 (72.4%)									
Normal	1 (3.4%)	2 (6.9%)	3 (10.3%)	3 (10.3%)									
High	12 (41.4%)	4 (12.8%)	5 (17.2%)	5 (17.2%)									

Note. χ^2: p-value for the Chi-Square test; κ: interclass coefficient.

a significant difference (p<0.0001) for the subcutaneous adiposity classification. All fixed-landmarks showed coefficients of agreement below the cut-off point (κ≥0.8) for the percentile classification of subcutaneous adiposity (κ<0.759) and normative relative body fat (κ<0.075).

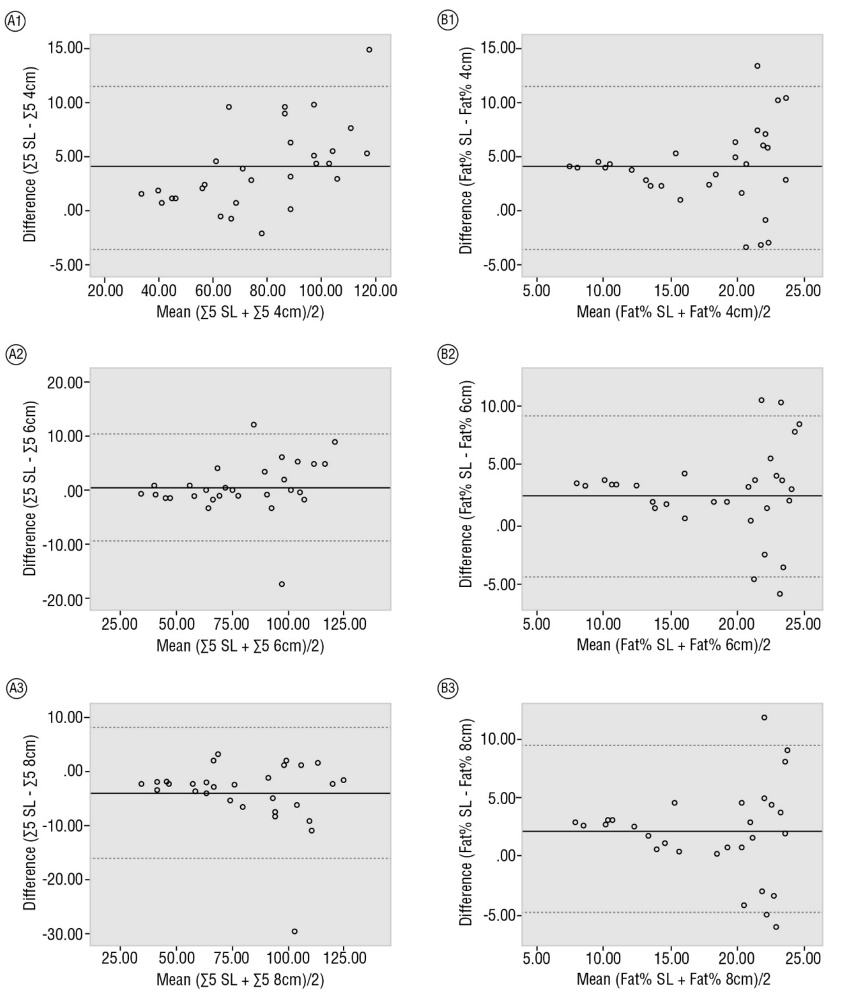

Figure 2. Bland-Altman plots for subcutaneous adiposity (A) and relative body fat (B) between the subjective-landmark and the fixed-landmarks of 4 cm (A1 and B1), 6 cm (A2 and B2) and 8 cm (A3 and B3).

DISCUSSION

The international protocol for anthropometric measurement is from a technical point of view periodically revised[19] in justification for the continuous updating of the literature. Comparative studies have investigated the reading time of skinfold thickness measurement[20], the physical-mechanical characteristics of the main types of skinfold calipers[21], interchangeable anthropometric measurement approaches[5,12,22] and the location of the skinfold site[5]. A study carried out with 62 male subjects observed that variation in the depth position of the skinfold caliper contact jaws produced significant differences in triceps skinfold thickness (p<0.05). The deep position resulted in thicker measurements and the superficial position resulted in less thick measurements, when compared to the middle position[4]. Burkinshaw et al.[23] found that marking the site of the four skinfolds in advance allowed examiners of varying degrees of technical skill to obtain consistent measurements. Subsequently, the importance of accurately locating the anatomical site of eight internationally standardized skinfolds was investigated. Hume and Marfell-Jones[5] observed in a sample of 10 male subjects that measuring 1 cm away from a site defined by the ISAK produced significant differences in most of the obtained skinfold measurement values.

The lack of analysis of the influence of measurement technique in the assessment of body composition or nutritional status is a methodological limitation of some comparative studies[4,5,12,23]. Outcome classification is an important guiding variable for prescriptive interventions. The present study quantified the effect of different pinch sizes on the thickness of eight internationally standardized skinfolds in a sample of 29 subjects, totaling more than 1.800 points of morphological data, and on the consequent interference in the estimation and classification of body adiposity components. The 6 cm-landmark showed similarity and statistical agreement with the subjective-landmark for all skinfolds, except the thigh, and with the sums of five and eight skinfolds. The 4 cm-landmark showed statistical similarity with subjective-landmark for triceps, supraspinale and calf skinfolds, however, there was agreement only for appendicular skinfolds. The 8 cm-landmark showed similarity and statistical agreement with subjective-landmark supraspinale and abdominal skinfolds. Thus, it appears that skinfold thickness pinching at limb sites needs to be a smaller size (<6 cm), except the thigh (>6 cm), and trunk sites needs to be a larger size (>6 cm). This evidence can have useful practical implications when the standardization of a fixed-landmark becomes necessary. The suggested opposite size between the lower appendicular sites is trivial. Martin et al.[3], in experiments with cadavers, it was evidenced that, regardless of gender, the thickness of the skin of the thigh is greater than that of the calf. Also, the static compressibility of the thigh is lower. In addition, the characteristic muscle volume of this segment implies greater skin resistance to pinching, especially in subjects undergoing strength training.

The measurement variation observed between the anatomical regions and the landmarks (Table 1) can be explained by the inverse relationship between the density and compressibility of the subcutaneous adipose tissue at each site[3,21]. Therefore, a high tissue density skinfold is less compressible compared to a low tissue density skinfold. Pinching with subjective distance between the fingers is the one that best suits the biological variability of skinfold thickness and, in view of this, standardization of a fixed size of pinching seems to be improbable. And further, add to this the fact that, as described in Esparza-Ros et al.[6], the marking of the iliac crest skinfold site is performed from the technical-palpatory subjectivity with the subcutaneous tissue, making this parameter applicable to all other sites. It is suggested that the fixed-landmarks examined in the present study are not interchangeable for the measurement of skinfold thickness. In addition, systematic interference of the pinch size was observed for the estimation (Table 1) and classification (Table 3) of body adiposity components. Subcutaneous adiposity classification differed significantly ($p<0.0001$) and body fat classification was the least affected by the size of skinfold thickness pinching. However, regardless of the classification criteria, there was no agreement between the subjective-landmark and the fixed-landmarks (Table 3). When the measurement of skinfolds is not performed correctly, the potential error is inflated, making the absolute values and estimates of the molecular and tissue component of body adiposity questionable and not applicable[24]. Therefore, we reinforce the need to standardize the measurement technique and carry out supervised training with experienced instructors.

Access to the main skinfold measurement protocols is limited, especially in Latin American countries, as such protocols are described in book chapters that have not been revised in the 21st century[7,9] or that require participation in an accreditation course technical for-profit[6,19]. The most relevant information from the reference literature[6,7,9,25] was compiled to facilitate reproduction by researchers and health professionals who use surface anthropometry in different fields of application. These technical procedures have been revised, improved and operationally categorized into two steps: marking and measurement. All of which are sequentially performed on the right side of the body. The left hand should be used to pinch the site and the right hand to handle the skinfold caliper regardless of the anthropometrist's lateral dominance. The use of anthropometric tape and a dermographic pen are essential for the marking stage. We suggest the use of a calibrated skinfold caliper that has been developed according to the physical, mechanical and functional characteristics proposed by Edwards et al.[26]. A minimum of two sequential measurements should be taken at each skinfold site. The mean value is used. In the event of a TEM of >5%, a triplicate is performed and the intermediate value used for the site that presents this variation.

Marking: I) Identify and accurately mark the skinfold site; II) Mark the line of the vertical, diagonal or horizontal anatomical axis of the skinfold and a perpendicular line forming an intersection; III) In the direction of the anatomical axis, mark a short guideline for the position of the caliper jaws at 1 cm from the site. [Note: this line ensures that the jaws are positioned in the same location in repeated measures.]; IV) Perform some pinching on the site with the left phalanges of the index finger and thumb flexed, perpendicular to the anatomical axis, in order to become familiar with the skin and subcutaneous adipose tissue; V) Define a skinfold size in which two parallel layers of tissue come together without excessively stretching the skin in the external region of the pinch and finish by marking this subjectivity with two points. *Measurement*: I) Position your fingertips on the pinching guide, then firmly detach a skinfold, with the back of the hand facing the anthropometrist, just above the intersection and perpendicular to the anatomical axis; II) Apply the caliper jaws at the pre-marked distance of 1 cm and at median depth proportionally to the middle of the fingernail. [Note: this depth is also understood as the alignment between the distal interphalangeal curve of the thumb and the curve of the caliper rods.]; III) Carefully observe the dial indicator and then gradually release the caliper trigger, keeping the skinfold firmly held; IV) The measurement reading should be recorded within the 3rd second after releasing the caliper trigger to obtain the subcutaneous adipose tissue static compressibility plateau; V) Remove the jaws by activating the caliper trigger and then release the skinfold.

This study involved intentional sampling and not representative of the morphological heterogeneity inherent in the population investigated. Therefore, the results are limited, in their ability to generalize, to groups with different characteristics of skinfold composition and compressibility. In addition, the lack of statistical analysis stratified by gender, which consequently limits the understanding of the results regarding sexual dimorphism. Our experimental evidence is important to update the internationally standardized skinfold measurement technique. It is recommended that the anthropometrist define and mark the size of the subjective distance between the fingers. Thus, the same skinfold thickness can be pinched in duplicate, which can increase the degree of intra-evaluator reproducibility.

CONCLUSION

Variation in pinch size is an important source of TEM that can affect the reproducibility of skinfold thickness measurements and interfere in the estimation and classification of the molecular and tissue component of body adiposity in a sample of adults.

COMPLIANCE WITH ETHICAL STANDARDS

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Ethical approval

The study followed the Brazil's National Health Council's research guidelines involving human experimentation. Approval was obtained by the Ethics and Research Committee of Platform Brazil under the University of Fortaleza, with number: CAAE - 89306918.9.0000.5052

Conflict of interest statement

The authors have no conflict of interests to declare.

Author Contributions

Conceived and designed experiments: JHCA; Performed experiments: JHCA; Analyzed data: JHCA, FBO, MIFC; Contributed with reagents/materials/analysis tools: JHCA, FBO, MIFC, RFC, WLR; Wrote the paper: JHCA. All authors read and approved the final version of the manuscript.

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