Original Research

Comparison of Methods to Assess Quadriceps Muscle Volume Using Magnetic Resonance Imaging

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Purpose: To compare the precision of four methods to estimate the volume of quadriceps muscles using axial MRI.

Materials and Methods: Entire legs of 10 healthy young subjects were scanned using a 1.5 Tesla magnetic resonance imaging scanner and 4-mm-thick sections without any gaps. Quadriceps muscles were outlined on all of the slices to obtain the MRI reference standard measure of quadriceps muscle volume. This MRI reference standard was compared with the volume estimated using (i) the truncated cone formula, (ii) the Cavalieri method, (iii) a cubic spline interpolation of missing cross sectional areas, and, (iv) the deformation of a parametric specific object. For each method, 3 to 21 slices were used.

Results: The average volume error was significantly ($P < 0.001$) different in comparing the four methods (4.4%, 2.3%, 1.1%, and 1.2%, respectively). In addition, the number of slices required to reach a given volume error was significantly ($P < 0.001$) different across all methods (respectively, 12, 9, 5, and 7 slices required to reach a volume error of 1.1%).

Conclusion: While methods based on interpolation and deformation of a parametric specific object have not been used in literature, these two methods are the most precise approaches to reach a given level of precision.

Key Words: muscle cross sectional area; Cavalieri formula; interpolation; truncated cone
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THE ESTIMATION OF muscle size is essential to assess hypertrophy or atrophy induced by strength training (1–6), intensive sport practice (7), aging (8–10), immobilization (11,12), space flight (13,14), or neuromuscular pathologies (15,16). The evaluation of muscle volume (MV) is based on several methods including MRI (5,6,17–19), computed tomography (CT) (20,21), ultrasonography (22,23), or bioelectrical impedance (24,25). In all of these studies, the reference standard measure used to validate MV estimations is obtained using axial MRI or CT. Because semiautomated or fully automated segmentation methods of MRI or CT images are inefficient because muscle distinction is often difficult or impossible to perform (Jolivet et al., 2008), MV estimations require practitioners to manually outline the muscles on several images to assess their cross-sectional area (CSA). However, this process is time consuming. For example, Tracy et al (6) reported that 370 h of analysis time were required to measure three replicates of quadriceps MV for both legs of 42 subjects. It probably explains why many studies have provided a single CSA measurement to assess muscle size (7,26).

Nevertheless, because it has been shown that a single CSA measurement is not necessarily representative of quadriceps MV changes (6,27), there is a need for a fast and precise method of measurement of MV. Moreover, the number of slices used in literature to assess quadriceps MV using axial MRI is very variable. It is obvious that the measurement error is increased when the number of available slices used to assess MV is decreased, yet only two studies have assessed the measurement error associated with the number of slices used to calculate MV. When reducing the number of slices from 50 to 8, Lund et al (18) showed that the error is negligible for ankle dorsi flexors (*tibialis anterior*, *extensor digitorum longus*, and *extensor hallucis longus*). Nevertheless, the measurement error was approximately 15% when using four slices. Tracy et al (6) have determined quadriceps MV using sections with 2-, 4-, 6-, 8-, 10-cm intervals. They considered that using a 4-cm interval, representing approximately 11–12 slices, provides a volume measurement with an accuracy of approximately 99%.

To estimate the MV using a given number of CSA measurements, a suitable calculation method must be selected. Several methods could then be used to esti-

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mate MV using a given number of CSA measurements. However, the choice of the method used can greatly influence the precision of the MV estimation. First, the truncated cone formula has been used to estimate MV (6,18). Lund et al (18) have reported no significant differences between the truncated cone formula and the Cavalieri approximation assuming an ideal cylinder between each slice. Shen et al (28) also reported similar volume measurements of human tissue and organ volumes, but the Cavalieri formula showed a slightly lower error in the volume estimation. However, an evaluation of the relevant literature reveals at least two other methods that could be used to assess MV using a reduced number of axial slices. Morse et al (19) have used third-order polynomial regressions on the relationship between quadriceps muscle CSA and the sections' distance to the patella to assess MV knowing only one CSA measurement and the muscle length. Nevertheless, the accuracy of this method was approximately 90%, and it requires knowing regression equations for the considered population. To our knowledge, no previous study has used an interpolation on a higher number of slices to improve the precision of this method. In addition, Jolivet et al (29) have recently developed a specific method, based on the deformation of a parametric specific object (DPSO), to assess muscle geometry using a reduced number of axial images. It consists of generating approximate parametric ellipses using basic dimensions of muscle contours, and building a regular surface mesh using an interpolation of the ellipses. This approximate object and the corresponding mesh are deformed to fit the exact muscle contours yielding subject-specific geometry. Jolivet et al (29) have shown that by using this method, the volume and the shape of the *gluteus maximus*, *gluteus medius*, *gluteus minimus*, *fascia lata tensor*, and *sartorius* muscles could be obtained with a satisfactory accuracy using approximately 5–6 slices (averaged volume error of 2.4%).

To our knowledge, four methods could be used to assess MV, but the precision of these methods has never been compared. Therefore, the objective of this study was to compare the precision of each method to assess quadriceps MV using MRI as a function of the number of slices. The outcomes of this study would provide important information about the recommended method(s), and the minimal number of slices needed to reach the required precision.

MATERIALS AND METHODS

Subjects

Ten healthy young males (29 ± 4 years; range, 24–34 years; height, 177.4 ± 6.1 cm; range, 168–185 cm; weight, 74.2 ± 7.5 kg; range, 65.7–91.5 kg; body mass index: 23.8 ± 1.8 $kg.m^{-2}$; range, 21.1–26.6 $kg.m^{-2}$) volunteered to participate in this study and signed an informed consent form. This study was conducted according to the Helsinki Statement (1964) and with the approval of the local ethical committees.

MRI

A Siemens Avanto 1.5 Tesla MRI scanner (Siemens, Munich, Germany) was used to obtain 3 series of axial images from the iliac spines to the calcaneum. The VIBE (Volume Interpolated GRE) sequence was chosen to obtain good quality images of the muscles of the lower limb in a reasonable time (6 min per series). An angiography RF coil (Siemens, Munich, Germany) was used so as to encompass the entire lower limb. For each series, parameters included repetition time = 4.47 ms; echo time = 2.10 ms; field of view: 400 × 400 mm; and voxel size =0.78 × 0.78 × 4 mm. Slice thickness was 4 mm without interslice interval.

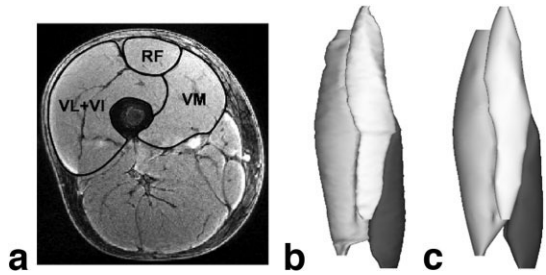

Figure 1. a: Typical example of axial image where *vastus lateralis* and *vastus intermedius* (VL+VI), *rectus femoris* (RF), *vastus medialis* (VM) muscles were manually outlined using the graphical interface developed in our laboratory. **b:** Typical example of three-dimensional reconstructions of VL+VI (medium shading), RF (light shading) and VM (dark shading) muscles generated with all the outlined slices and used to calculate the MRI reference standard measure of quadriceps volume. **c:** Typical example of three-dimensional reconstructions of VL+VI (medium shading), RF (light shading), and VM (dark shading) muscles using the Deformation of a Parametric Specific Object method and 7 available slices.

The *vastus lateralis* (VL), *vastus intermedius* (VI), *vastus medialis* (VM), and *rectus femoris* (RF) muscles of the 10 subjects were manually outlined for all of the slices by the same investigator using a graphical interface implemented by using Matlab® (The Mathworks, Natick, MA), as developed in our Laboratory (Fig. 1A). Because substantial fusion may be found between VL and VI on some slices (3), these two muscles were outlined together.

Methods to Assess MV

All of the slices (90 to 107 slices depending on each subject's thigh length) were first outlined by an investigator, from the most distal slice where quadriceps muscle can be seen, to the most proximal slice. Using all of the outlined slices, the three-dimensional shapes of the three muscles (VL+VI, VM, RF) were generated (Fig. 1B) to determine the MRI reference standard measure of MV (29). For this procedure, each muscle contour was discretized using N points regularly spaced, then each contour was connected to each other to construct a surface constituted of triangular polygons. Quadriceps MV was then calculated as the sum of these three MVs. Muscle CSAs were also calculated for each slice. The MRI reference standard measures of MV were compared with the MV calculated using four methods, and considering different numbers of available slices. A pilot study has shown that the methods converge relatively rapidly, so we chose to vary the number of avail-

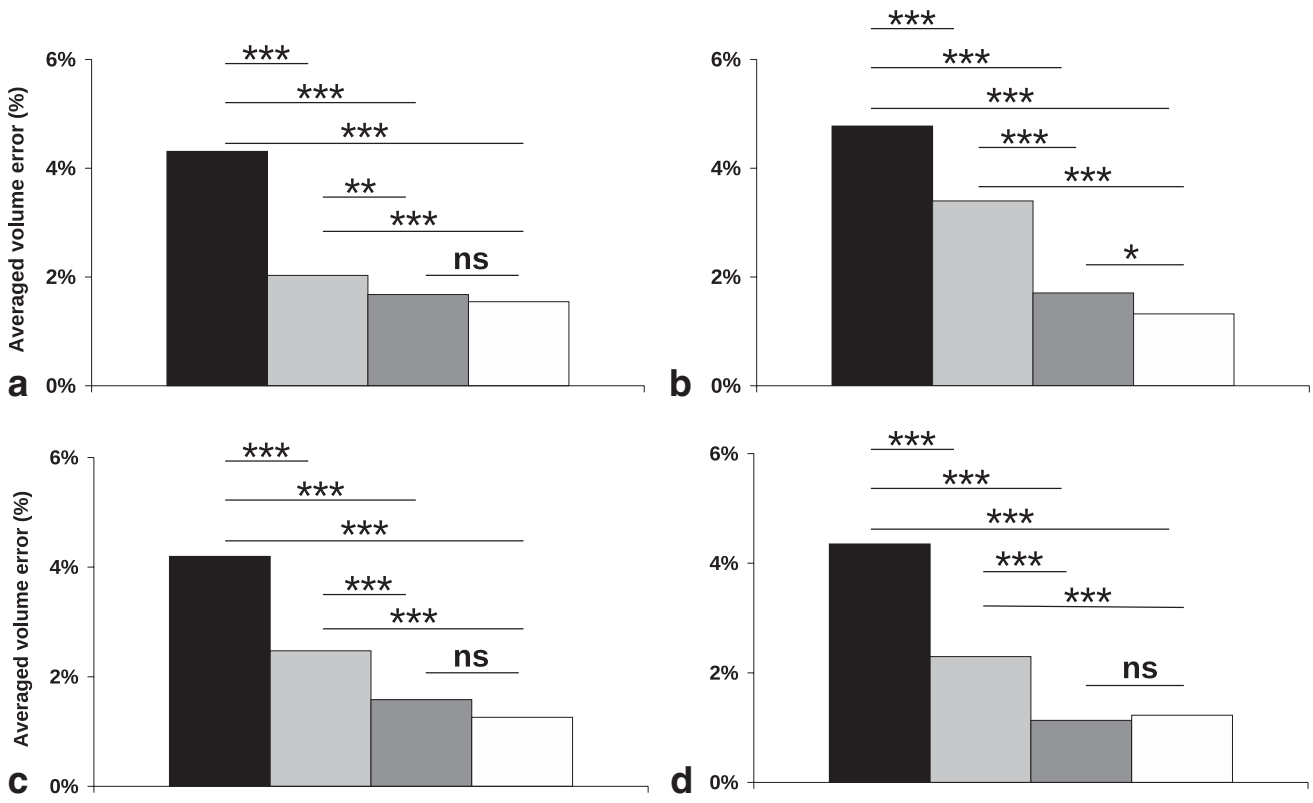

Figure 2. a–d: Average error of *vastus lateralis* + *vastus intermedius* (a), *vastus medialis* (b), *rectus femoris* (c), and quadriceps (d) muscle volume estimations using the truncated cones (black shading), Cavalieri (light shading), interpolation (medium shading), deformation of parametric specific objects (white) methods in respect to the MRI reference standard measure. The significant main effect ($P < 0.001$) of method in the 4 × 19 (method × number of slices) ANOVA is presented. *$P < 0.05$; **$P < 0.01$; ***$P < 0.001$; ns: nonsignificant

able slices to estimate MV from 3 to 21. The four considered methods were as follows.

- The truncated cone formula [1] (30), which assumes a conical shape of the muscle between available slices:

$$MV = \sum_{n-1} \frac{e_i}{3} \times (CSA_i + CSA_{i+1} + \sqrt{CSA_i \times CSA_{i+1}}) \quad [1]$$

  Where n is the number of slices used, and e_i is the distance between available slices i and i+1
- The Cavalieri formula [2] (18), which assumes an ideal cylinder shape of the muscle between the available slices

$$MV = \sum_{n} e_i \times CSA_i \quad [2]$$

- A cubic spline interpolation was used for the relationship between CSA calculated on available slices and distance from insertion to estimate missing CSAs. The MV was then calculated using the Cavalieri formula [2].
- The deformation of a parametric specific object (DPSO) method (29), which could be decomposed in four steps. (i) Each muscle contour obtained on available slices was modeled as an equivalent ellipse. The centroid coordinates, local inertial coordinate systems, width and length were calculated for each available ellipse. (ii) Changes in these parameters along the muscle's principal axis were modeled using the cubic spline interpolation to estimate missing ellipses. Using all available and estimated ellipses, the resulting three-dimensional parametric object was reconstructed. (iii) The subject-specific volumetric muscle reconstruction (Fig. 1C) was determined by deforming the parametric object using a nonisotropic algorithm (31) and available muscle contours. (iv) The MV calculation was performed on this three-dimensional volumetric muscle reconstruction.

Intra- and Inter-Investigator Reproducibility of the MRI Reference Standard Measure

The precision required for the MV depends on the reproducibility of the MRI reference standard measure obtained with exhaustive muscle contours. Therefore, all of the slices were outlined a second time on different days by the same investigator, and on another occasion by a second investigator to assess the intra- and inter-investigator reproducibility of our MRI reference standard measure.

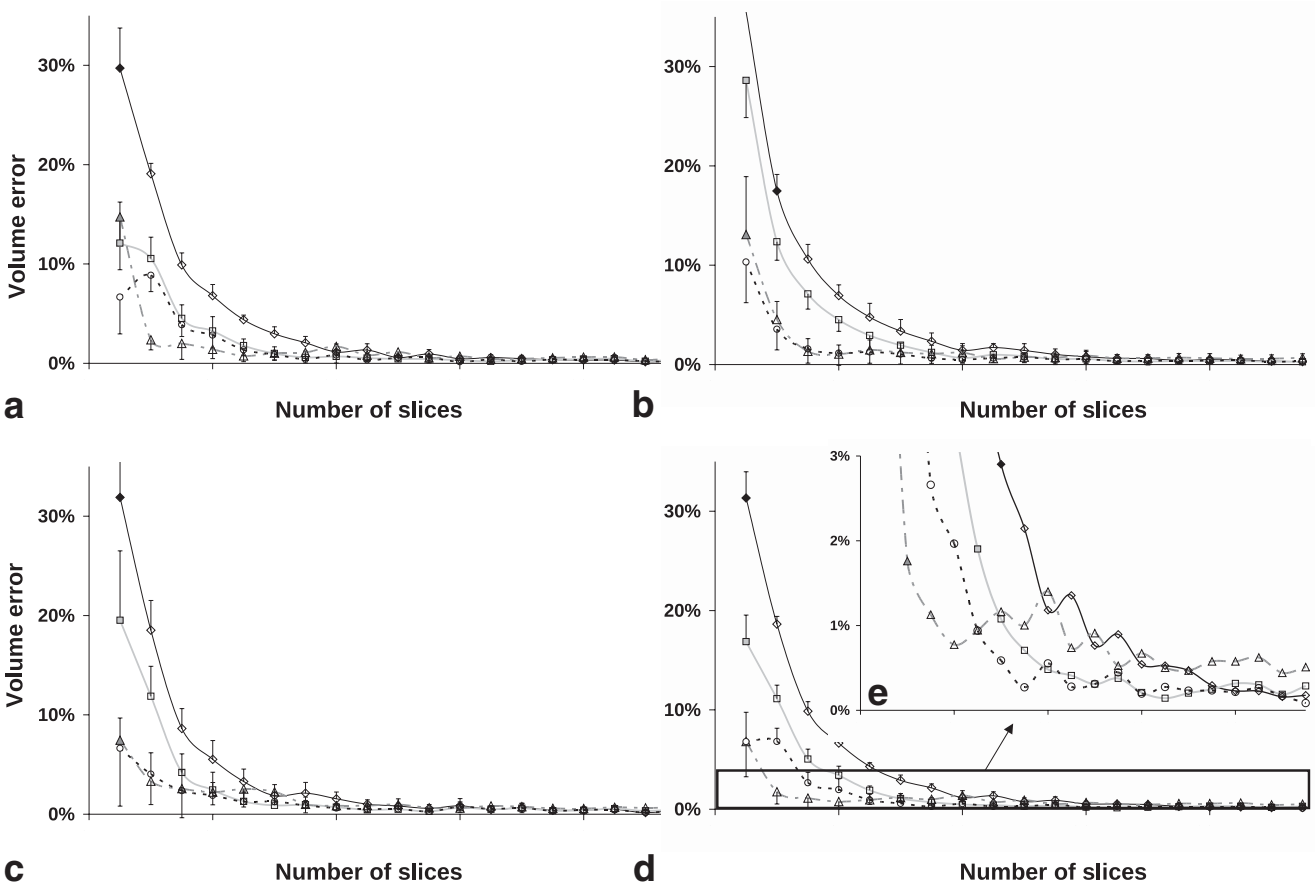

Figure 3. a–d: Error of *vastus lateralis* + *vastus intermedius* (a), *vastus medialis* (b), *rectus femoris* (c), and quadriceps (d) volume estimations using the truncated cones (diamonds), Cavalieri (squares), interpolation (triangles), deformation of parametric specific objects (circles) methods as functions of the number of available slices in respect to the MRI reference standard measure. The significant main interaction ($P < 0.001$) of the 4 × 19 (method × number of slices) ANOVA is described. **e:** A detail of (d).

Statistical Analysis

Intra- and interinvestigator reproducibilities were determined using the intraclass correlation coefficient (ICC) and the coefficient of variation (CV) (32) for the quadriceps MV. The global reproducibility of the MRI reference standard quadriceps MV measure was also evaluated, after checking that the differences were normally distributed (Shapiro-Wilk test), as 1.96 SD of the differences between the mean and the three MV measurements.

The volume error was determined for each method as the normalized absolute difference between MV estimated for VL+VI, VM, RF, and quadriceps and the MRI reference standard measures of MV for all numbers of available slices (33). After checking the normality of data distribution (Shapiro-Wilk test), four 4 × 19 (method × number of available slices to estimate MV) analysis of variance (ANOVA) were used to compare the volume error between the methods for VL+VI, VM, RF, and quadriceps MV. The critical level of significance in the present study was set at $P < 0.05$. Least significant difference post hoc analysis was used when appropriate. In addition, the bias and the limit of agreement (1.96 SD) between the MRI reference standard measure of quadriceps MV and MV estimated using the four methods were determined using Bland-Altman plots (33,34). Paired t-tests were performed for each Bland-Altman plot to determine whether the bias was significantly different from zero.

RESULTS

Intra- and Interinvestigator Reproducibility of the MRI Reference Standard MV Measure

The mean MVs for the three measurements for the quadriceps were 2430 ± 399 cm^3 (first investigator), 2432 ± 393 cm^3 (first investigator), and 2419 ± 398 cm^3 (second investigator). Excellent intra- and interinvestigator reproducibility was observed (ICC = 0.999 and 0.997, respectively; CV = 0.5% and 0.8%, respectively). The global reproducibility was 1.1% (27 cm^3).

Accuracy of Methods

The significant effect of the number of slices ($P < 0.001$) confirmed that the volume error was decreased when the number of available slices was increased for VL+VI, VM, RF, and quadriceps muscles. A significant main effect of the method ($P < 0.001$) showed that the volume errors of the four methods were different (Fig. 2). For the

Table 1
Bias and Limits of Agreement (1.96 SD) for the Four Considered Methods of Quadriceps Muscle Volume Estimation Compared to the MRI Reference Standard Measure

No. of slices	Truncated cones					Cavalieri					Interpolation					DPSO				
	Bias		T[a]	1.96 SD		Bias		T[a]	1.96 SD		Bias		T[a]	1.96 SD		Bias		T[a]	1.96 SD	
	(cm^3)	(%)		(cm^3)	(%)	(cm^3)	(%)		(cm^3)	(%)	(cm^3)	(%)		(cm^3)	(%)	(cm^3)	(%)		(cm^3)	(%)
3	763	37.3	***	272	7.2	410	18.5	***	171	6.2	−166	−6.5	***	173	6.5	168	7.1	***	148	6.2
4	453	20.5	***	168	1.9	271	11.8	***	124	3.0	36	1.5	*	85	3.3	168	7.1	***	106	2.8
5	239	10.4	***	78	2.1	122	5.2	***	50	2.1	22	1.0	*	55	2.3	65	2.7	***	52	2.1
6	163	6.9	***	62	1.7	83	3.5	***	48	1.9	5	0.2	ns	47	1.9	48	2.0	***	55	2.2
7	105	4.4	***	35	0.9	46	1.9	***	22	0.9	−24	−0.9	***	29	0.9	23	0.9	***	27	1.1
8	74	3.1	***	42	1.1	27	1.1	***	29	1.0	−27	−1.1	***	27	1.1	12	0.5	*	36	1.4
9	52	2.2	***	17	0.8	17	0.7	***	19	0.9	−25	−1.0	***	25	0.9	5	0.2	**	12	0.5
10	29	1.2	***	37	1.5	1	0.0	ns	30	1.3	−33	−1.4	***	32	1.4	−6	−0.3	ns	32	1.4
11	33	1.4	***	19	0.7	10	0.4	**	19	0.8	−17	−0.7	***	22	0.9	5	0.2	ns	13	0.6
12	17	0.7	***	19	0.7	0	0.0	ns	17	0.7	−22	−0.9	***	20	0.8	−2	−0.1	ns	18	0.8
13	21	0.9	***	19	0.8	5	0.2	ns	17	0.7	−13	−0.5	***	22	0.9	5	0.2	ns	22	0.9
14	14	0.6	***	16	0.7	−1	0.0	ns	11	0.5	−16	−0.7	***	13	0.5	1	0.0	ns	11	0.5
15	12	0.5	***	10	0.3	1	0.0	ns	10	0.4	−12	−0.5	***	11	0.4	3	0.1	ns	15	0.6
16	9	0.4	**	16	0.7	0	0.0	ns	15	0.6	−11	−0.5	***	14	0.6	3	0.1	ns	12	0.5
17	5	0.2	*	12	0.5	−5	−0.2	*	14	0.6	−14	−0.6	***	15	0.5	−1	0.0	ns	15	0.6
18	3	0.1	ns	13	0.6	−6	−0.2	*	14	0.6	−14	−0.6	***	17	0.6	0	0.0	ns	13	0.6
19	2	0.1	ns	13	0.6	−7	−0.3	**	12	0.5	−15	−0.6	***	12	0.5	−2	−0.1	ns	14	0.6
20	3	0.2	*	11	0.5	−4	−0.1	*	9	0.3	−11	−0.4	***	10	0.3	1	0.0	ns	12	0.5
21	1	0.0	ns	14	0.6	−5	−0.2	*	12	0.5	−12	−0.5	***	12	0.5	1	0.0	ns	6	0.3

[a]Student t-test used to determine if the bias is significantly different from zero.
*$P < 0.05$;
**$P < 0.01$;
***$P < 0.001$.
ns = non significant; DPSO = deformation of parametric specific objects method.

quadriceps MV, the volume error averaged across the numbers of slices was 4.4% for truncated cones method, 2.3% for Cavalieri method, 1.1% for interpolation method, and 1.2% for DPSO method, respectively. Finally, the significant interaction of method × number of available slices ($P < 0.001$) is shown in Figure 3. It indicates that the number of slices required to reach a given volume error was significantly different for the methods compared. Similar results were obtained for VL+VI, VM, RF, and quadriceps muscles (Figs. 2, 3). For the quadriceps MV, the volume error of the truncated cone method was significantly ($P < 0.05$) higher than the volume error of the Cavalieri and DPSO methods from 3 to 11 slices. The truncated cones method was also significantly ($P < 0.05$) higher than the volume error of the interpolation method from 3 to 9 slices. The volume error of the Cavalieri method was significantly ($P < 0.05$) higher than the volume error of the interpolation and DPSO methods from 3 to 7 slices. The error of the interpolation method was significantly ($P < 0.05$) lower than the method of the DPSO method from 4 to 9 slices, while it was significantly higher for 9 and 10 slices.

The bias and the limits of agreement (1.96 SD) associated with each method and numbers of slices for quadriceps MV are shown in Table 1. The bias was significantly different from zero except for 18, 19 and 21 slices, using the truncated cone method; 12 to 16 slices using the Cavalieri method; 6 slices using the interpolation method; and, 10 to 21 slices using the DPSO method, respectively.

Considering the reliability of the MRI reference standard for the quadriceps MV, the number of slices required to reach a volume error of 27 cm^3 (i.e. 1.1%) was analyzed. Thus, a volume error of 27 cm^3 resulted in 12, 9, 5, and 7 slices for truncated cones, Cavalieri, interpolation, and DPSO methods, respectively. Bland-Altman plots showing the bias and the limits of agreement for the MV estimated using these numbers of slices are shown Figure 4.

DISCUSSION

The present study was designed to compare four methods to estimate quadriceps MV using MRI. These estimations of MV were compared with the MRI reference standard measure performed using all of the slices. To obtain this MRI reference standard, quadriceps muscles were outlined on approximately 100 slices, which required more than 90 min of analysis time per subject. Concerning this MRI reference standard measure, our results show excellent intra- and interinvestigator reproducibility with a global reproducibility of 1.1%. In addition, similar errors were found for the three considered muscles (VL+VI, VM, RF, and quadriceps) (Figs. 2, 3), indicating that the error associated with each method does not seem to depend on the muscle shape. Consequently, we focus the discussion on the quadriceps MV calculations.

Results of the present study show that the analysis time could be reduced to less than 15 min with one of the four considered methods that require analyzing

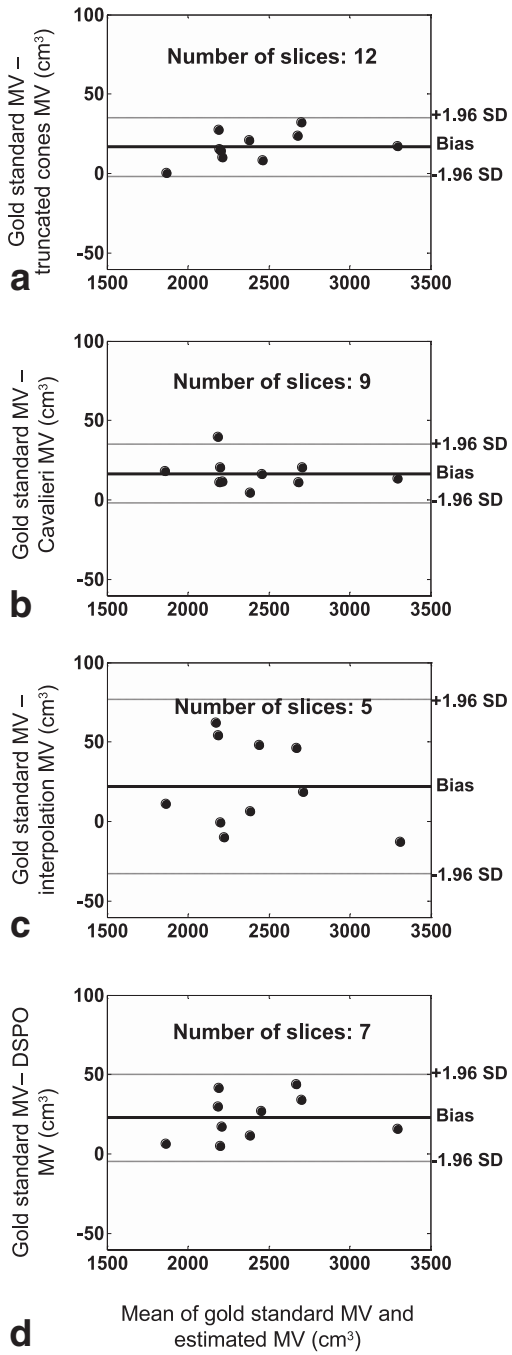

Figure 4. Examples of Bland-Altman plots showing the bias and the limits of agreement (± 1.96 standard deviation [SD]) for quadriceps muscle volume (MV) estimated using the smallest number of slices required to reach a volume error lower than 1%. **a:** Truncated cones method. **b:** Cavalieri method. **c:** Interpolation method. **d:** Deformation of a parametric specific object (DPSO) method. The bias was significantly different from zero for a–d.

fewer slices. The volume error (Fig. 3), bias, and limits of agreement (Table 1) were significantly different across the methods tested. Thus, the truncated cones method, which seems to be the most often used method reported in literature (5,6,18,30,35), is also significantly less precise in the estimation of quadriceps MV, and should not be used when less than 12 slices are used. This result is not in accordance with the study of Lund et al (18) who have reported no significant differences between the truncated cones and Cavalieri methods. The statistical analysis conducted by Lund et al was performed on 50, 34, 26, 20, 18, 14, 12, 10, 8, and 4 slices (18). Therefore, the number of slices was in general higher than in the present study, inducing lower global errors. The lower number of slices in the study of Lund et al (18) probably smoothed the differences between methods and explains the discrepancies with results of the present study. Because the Cavalieri method can be considered as an approximation of the truncated cones method, it seems surprising that the truncated cones method is less precise. However, this result is in accordance with a previous study concerning the volume assessment of various human tissue and organs (28), and it could be explained by using Figure 5, which shows a typical example of the relationship between the distance from the patella and the CSA of VL + VI assessed by using the different methods. In fact, the Cavalieri method is the less precise method to estimate the shape of this relationship, but cumulative errors in the CSA estimation cancel each other out. In contrast, the truncated cones method leads to a satisfactory estimation of the shape, but also to a constant underestimation of the CSA, and hence to the quadriceps MV.

The method and the number of slices that should be selected depend on the precision needed for a given study. Thus, future investigators could choose the appropriate method using results reported herein, in Figure 3 and Table 1. Using the truncated cone formula for young men, Tracy et al (6) obtained biases of 23, 67, 117, and 170 cm^3 for 4, 6, 8, and 10 cm of intervals between slices, respectively. Considering a thigh length of approximately 40–45 cm, it would require outlining, respectively, approximately 11–12, 8, 6, and 5 slices. Therefore, these results are similar to those reported in Table 1 showing biases of 17, 74, 163, 239 cm^3 for 12, 8, 6, and 5 slices, respectively. Finally, Tracy et al (6) have shown that quadriceps MV is estimated with a

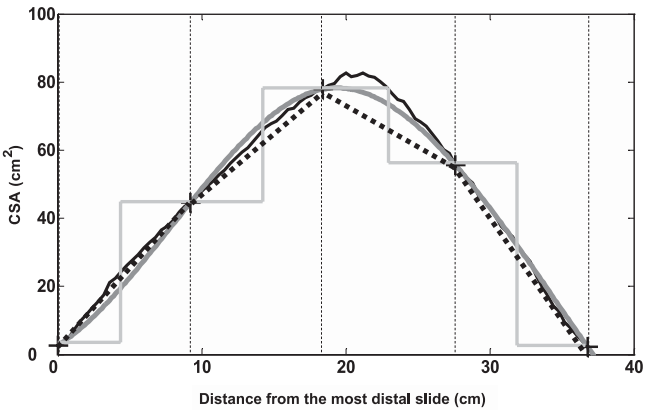

Figure 5. Cross-sectional area (CSA) of *vastus lateralis* + *vastus intermedius* muscle as a function of the distance from the most distal slide used to assess volume using all the slices. CSA estimated using truncated cones (dotted line), Cavalieri (light shaded line), and interpolation (dark shaded line) methods with five available slices and the MRI reference standard measure (black line).

satisfactory accuracy (~99%) using a 4-cm interval between slices.

Taking into account the reliability of the MRI reference standard, an error of quadriceps muscle MV estimation of less than 27 cm^3 (1.1%) can be considered meaningless. Hence, we also determined the number of slices required to reach this volume error, which was 12, 9, 5, and 7 using truncated cones, Cavalieri, interpolation and DPSO methods, respectively (Fig. 4). Tracy et al (5,6) have shown that using a 4-cm interval between slices and the truncated cone formula, the analysis time could drop from 370 to 90 h. Consequently, using the interpolation or DPSO methods rather than the truncated cone formula, it could drop to approximately 45 h with a similar accuracy. However, the interpolation method did not ever converge completely to 0% of volume error for the number of available slices considered in the present study (Fig. 3). Thus, a significant bias, sometimes higher that 1.1%, is present from 6 to 21 slices (Table 1). Consequently, if a high level of accuracy is needed, the DPSO method should be used.

If the DPSO method provides a precise measurement of quadriceps MV using a reduced number of slices, this method can also be used to determine muscle three-dimensional geometry, and to model the musculoskeletal system (Fig. 1C). Thus, Jolivet et al (29) reported a mean error between reconstructed and criterion objects of approximately 2 mm for *gluteus maximus*, *gluteus medius*, *gluteus minimus*, *fascia lata tensor*, and *sartorius* muscles. A preliminary study appears to show that this method could be efficient to assess muscle shape, line of action, and moment arm of quadriceps muscles, but these results need to be confirmed.

In conclusion, the present study shows that the quadriceps MV estimation is significantly different among the four methods tested herein. Based on these data, future investigators could then choose the ideal method and the number of slices depending on the specific precision required for their study. It must be emphasized that the truncated cone formula was the least accurate method. In addition, to reach a volume error of 1.1%, the interpolation and DPSO were the most precise of the methods tested. Finally, a precise and efficient quadriceps muscle volume calculation is performed using the DPSO method, which could also be used to assess muscle geometry. Further investigation is needed to determine the precision of this method to estimate muscle shape, line of action, and moment arm.

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