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Image J software differences in B-mode ultrasound imaging of the intrinsic plantar muscles cross-sectional area between hemiparetic and contralateral feet in post-stroke patients: A case-control study

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Image J software differences in B-mode ultrasound imaging of the intrinsic plantar muscles cross-sectional area between hemiparetic and contralateral feet in post-stroke patients: A case-control study

Abstract

Background: To date, the Image J software analysis of the intrinsic plantar muscles cross-sectional area (CSA) has not yet been analyzed in poststroke patients.

Objective: Thus, the aim of this study was to analyze and compare the Image J software differences in B-mode ultrasound imaging of the intrinsic plantar muscles CSA between hemiparetic and contralateral feet in poststroke patients.

Methods: A convenience sampling was carried out in order to recruit 22 feet from 11 poststroke patients. The total sample was divided into 11 hemiparetic feet (case group) and 11 contralateral feet (control group). The ImageJ software was utilized in order to measure the CSA as well as the mean, standard deviation (SD) and count of the pixels from all the offline images of the abductor hallucis, flexor digitorum brevis and flexor hallucis brevis.

Results: Statistically significant differences ($P = 0.003$) were only shown for the AbH pixels count. The rest of measurements did not show any statistically significant difference ($P > 0.05$).

Conclusion: Image J software differences in B-mode ultrasound imaging of the AbH pixels count were presented between hemiparetic and contralateral feet in poststroke patients. Based on the present study, future studies about advances in pixel analysis with Image J could help understand the relationship with neuromuscular alterations and musculoskeletal composition changes.

Keywords: Anatomy, Cross-sectional; Foot; Ultrasonography; Paresis.
Introduction

Strokes may be defined as a common syndrome that occurs in developed countries (prevalence 1.47 - 2.6%) secondary to alterations of the brain blood flow [1]. In Spain, an incidence up to 220 strokes per 100,000 individuals per year was reported [1,2]. Furthermore, a high mortality was shown [3]. The most frequent mortality age range varied from 38 to 50 years old [1].

In the European countries, high economic costs were shown secondary to stroke [4]. The characteristics and treatment may depend on the stroke types (i.e. hemorrhagic or ischemic). Strokes represented a major disability cause with neurologic disorders and impairments such as walking [1,4].

Plantar flexor muscles of poststroke patients were considered as a key focus of assessment and intervention [5–7]. Indeed, ultrasound imaging was applied to evaluate the medial gastrocnemius muscle with a good reliability and may be utilized for clinical assessment in poststroke patients [6]. Nevertheless, there is a lack of research studies regarding the rehabilitative ultrasound imaging (RUSI) evaluation of the intrinsic plantar muscles in poststroke survivors.

Despite a lack of research about ultrasound imaging in the intrinsic plantar muscles is shown in poststroke patients, prior studies assessed the cross sectional area (CSA) of these foot muscles, such as flexor digitorum brevis (FDB), flexor hallucis brevis (FHB) and abductor hallucis (AbH) with an excellent reliability [8–10].

Prior studies with different analysis image software were applied in order to evaluate B-mode ultrasound image from different musculoskeletal structures [11,12]. To date, the Image J software analysis of the intrinsic plantar muscles CSA has not yet been analyzed in poststroke patients [13]. Thus, aim of this study was to analyze and compare the Image J software...
differences in B-mode ultrasound imaging of the intrinsic plantar muscles CSA between hemiparetic and contralateral feet in poststroke patients.

Methods

Design

A case-control study was performed from June 2017 to January 2018. The Strengthening the Reporting of Observational Studies in Epidemiology (STROBE) statements were considered [14].

Sample size calculation

A sample size calculation was carried out by the difference between two independent groups thought the G*Power 3.1.9.2 software and based on the FDB pixels mean of a pilot study (n = 8) with 2 groups (mean ± SD), 4 feet with hemiparesis (81.75 ± 3.35 pixels) and 4 contralateral feet (69.81 ± 14.82 pixels). Furthermore, a 1-tailed hypothesis, an effect size of 1.11, an α-error probability of 0.05, a power (1-β error probability) of 0.80 and an allocation ratio (N2/N1) of 1 were used for the sample size calculation. Thus, a total sample size of 22 feet, 11 feet with hemiparesis and 11 contralateral feet, was provided.

Participants

A convenience sampling was carried out in order to recruit 22 feet from 11 poststroke patients at the poststroke unit of the Beata Maria Ana Hospital (Madrid, Spain). The total sample was divided into 11 hemiparetic feet (case group) and 11 contralateral feet (control group).

The inclusion criteria included individuals > 18 years without soreness in lower limb areas during the last 6 months [8,10]. The exclusion criteria included self-reported or medical record conditions (such as surgery, fracture, sprain, tear, tendinopathy, systemic alterations, and rheumatoid arthritis) [8–10,15].

Ethical approval
The Centro Superior de Estudios Universitarios La Salle Ethics committee (Madrid, Spain; code: CSEULS-PI-156/2017) evaluated and approved this research. Signed informed consent forms were collected before the study beginning. The human experimentation ethical guidelines and the Helsinki Declaration were respected [16].

**Descriptive data**

The descriptive data including sex (male or female), age (years), weight (kg), height (m), body mass index (BMI; kg/m$^2$), and foot side (right or left) were registered [8–10].

The ambulation capacity was assessed by an experienced neurology physician by means of the Functional Independence Measure and Functional Assessment Measure (FIM+FAM) with good psychometric properties of Spanish validation and reliability in poststroke patients [17–19]. The ambulation capacity was categorized in the following ranges: 1-total assistance, 2-maximum assistance, 3-moderate assistance, 4-minimum assistance, 5-supervision, 6-modified independency and 7-complete independency [17–20].

The Modified Modified Ashworth Scale (MMAS) was measured by an experienced neurology physician with a good agreement in order to evaluate the plantar flexor spasticity in poststroke patients. This scale was categorized in the following ranges: 0-normal muscle tone, 1-slight hypertonicity, 2-moderate hypertonicity, 3-high hypertonicity and 4-very high hypertonicity [21].

**B-mode ultrasound Imaging**

All ultrasound imaging measurements were performed by an expert physiotherapist with > 4 years of experience who was not blind to case or control group allocation. A high quality ultrasound tool (Ecube-i7; Alpinion-Medical Systems, Seoul, Korea) with a linear probe from 8- to 12.0-MHz-range (Broadband Linear type L3-12-T; 45-mm footprint was used to carry out resting B-mode ultrasound image.
The probe location was performed according to prior ultrasound imaging studies within intrinsic plantar muscles CSA measurements [8–10]. The CSA (perpendicular to the muscle fibers) was evaluated in the thickest portion of the AbH, FDB and FHB muscles in 3 different scanning lines. The AbH scanning line was placed from the navicular bone tuberosity to the medial calcaneus bone tuberosity. The FDB scanning line was located from the 3rd toe to the medial calcaneus tuberosity bone tubercle. The FHB scanning line was located along the 1st metatarsal shaft [8–10].

**Image J software analysis**

Finally, the Image J software analyses were carried out by an expert physiotherapist with > 4 years of experience (blinded to the case or control group allocation). The ImageJ software (version-2.0; US-National Institutes of Health; Bethesda-Maryland, USA) was utilized in order to measure the CSA as well as the mean, standard deviation (SD) and count of the pixels from all the offline images of the FDB (Figure 1), FHB (Figure 2) and AbH (Figure 3) [13]. The average of 3 repeated measurements was used for the data analysis. An excellent CSA reliability (intraclass correlation coefficient, ICC = 0.91 - 0.98) [8].
Figure 1. ImageJ software analysis of the CSA, mean, SD and count of the pixels from the FDB of the hemiplegic and contralateral feet in poststroke patients. Abbreviations: CSA, cross-sectional area; FDB, flexor digitorum brevis; SD, standard deviation.

Figure 2. ImageJ software analysis of the CSA, mean, SD and count of the pixels from the FHB of the hemiplegic and contralateral feet in poststroke patients. Abbreviations: CSA, cross-sectional area; FHB, flexor hallucis brevis; SD, standard deviation.
Figure 3. ImageJ software analysis of the CSA, mean, SD and count of the pixels from the AbH of the hemiplegic and contralateral feet in poststroke patients. Abbreviations: AbH, abductor hallucis; CSA, cross-sectional area; SD, standard deviation.

Statistical analysis

Statistical analysis was performed by the 22.0v SPSS (software IBM SPSS Statistics, Windows; Armonk-NY: IBM-Corp) considering an α error of 0.05 and a 95% confidence interval (CI) with a desired power of 80% (β error of 0.2) according to the sample size calculation.

Regarding the quantitative data, normality Shapiro-Wilk test was performed. Parametric data ($P > .05$) were described as mean ± standard deviation (SD) and range (minimum–maximum), and analyzed to compare descriptive data (weight and height) and all ultrasound and Image J measurements (CSA, mean, SD and count of pixels for FDB, FHB and AbH muscles) by the Student t test for independent samples. Non-parametric data ($P < .05$) were described as median ± interquartile range (IR) and rage (minimum–maximum), and
analyzed to compare descriptive data (age and BMI) by the Mann-Whitney U test. Considering the categorical data, frequencies and percentages were utilized to describe these data and the Chi-square ($\chi^2$) test was used to compare the MMAS and FIM+FAM data as well as Fisher exact test was used to compare the sex and foot side.

Results

Demographic and descriptive data

There were not statistically significant differences ($P > .05$) between hemiparesis and contralateral feet for sex (8 males / 14 females), age (median ± IR of 62.00 ± 5.00 years), weight (mean ± SD of 69.45 ± 17.32 kg), height (mean ± SD of 1.68 ± 0.11 kg), BMI (median ± IR of 19.55 ± 7.99 kg/cm$^2$), foot side (12 right / 10 left), MMAS (9 grades-II and 2 grades-III for the hemiplegic foot; 11 grades-0 for the contralateral feet) and FIM+FAM showing 0 (0%) grade-1, 4 (16.7%) grades-2, 8 (33.3%) grades-3, 4 (16.7%) grades-4, 5 (16.7%) grades-5, 2 (8.3%) grades-6 and 0 (0%) grades-7.

B-mode ultrasonography and Image J analysis of the intrinsic plantar muscles

The ultrasound measurements regarding the CSA, mean, SD and count of the pixels from the FDB, FHB and AbH of the hemiparetic and contralateral feet from poststroke patients were shown in Table 1. Regarding the intrinsic plantar muscles, statistically significant differences ($P = 0.003$) were only shown for the AbH pixels count. The rest of measurements did not show any statistically significant difference ($P > 0.05$).
Table 1. ImageJ software analysis of the CSA, mean, SD and count of the pixels from the intrinsic plantar muscles of the hemiplegic and contralateral feet in poststroke patients.

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Hemiparesis feet mean ± SD (n = 20)</th>
<th>Contralateral feet mean ± SD (n = 20)</th>
<th>P-value*</th>
</tr>
</thead>
<tbody>
<tr>
<td>CSA (cm²)</td>
<td>FDB 2.31 ± 0.58 (1.39–3.37)</td>
<td>2.57 ± 0.68 (1.34–3.43)</td>
<td>.341</td>
</tr>
<tr>
<td></td>
<td>FHB 2.08 ± 0.46 (1.11–3.86)</td>
<td>1.97 ± 0.45 (1.31–2.52)</td>
<td>.611</td>
</tr>
<tr>
<td></td>
<td>AbH 2.38 ± 0.56 (1.39–3.53)</td>
<td>2.78 ± 0.74 (1.84–4.42)</td>
<td>.167</td>
</tr>
<tr>
<td>Pixels count</td>
<td>FDB 65249.90 ± 18.089.52 (46028.00–95940.00)</td>
<td>83075.51 ± 23339.07 (39728.00–138092.67)</td>
<td>.102</td>
</tr>
<tr>
<td></td>
<td>FHB 62931.63 ± 11849.62 (48161.33–83519.33)</td>
<td>71732.91 ± 19720.70 (37901.00–103927.00)</td>
<td>.219</td>
</tr>
<tr>
<td></td>
<td>AbH 66572.48 ± 12770.22 (47111.00–85467.67)</td>
<td>87513.18 ± 15644.87 (57851.33–114668.00)</td>
<td>.003</td>
</tr>
<tr>
<td>Pixels mean</td>
<td>FDB 86.35 ± 10.59 (69.53–103.20)</td>
<td>75.57 ± 17.10 (47.72–111.09)</td>
<td>.091</td>
</tr>
<tr>
<td></td>
<td>FHB 88.19 ± 9.30 (71.39–99.70)</td>
<td>76.86 ± 15.62 (58.04–100.14)</td>
<td>.055</td>
</tr>
<tr>
<td></td>
<td>AbH 75.87 ± 12.81 (60.70–104.81)</td>
<td>67.13 ± 11.15 (60.70–104.81)</td>
<td>.107</td>
</tr>
<tr>
<td>Pixels DS</td>
<td>FDB 41.08 ± 5.90 (34.91–52.23)</td>
<td>38.57 ± 5.98 (29.69–49.77)</td>
<td>.334</td>
</tr>
<tr>
<td></td>
<td>FHB 33.04 ± 3.14 (28.81–36.50)</td>
<td>31.83 ± 5.79 (22.68–45.91)</td>
<td>.552</td>
</tr>
<tr>
<td></td>
<td>AbH 37.42 ± 0.25 (27.46–44.35)</td>
<td>38.05 ± 4.77 (31.39–47.06)</td>
<td>.760</td>
</tr>
</tbody>
</table>

Abbreviations: AbH, abductor hallucis; CI, confidence interval; CSA, cross-sectional area; FDB, flexor digitorum brevis; FHB, flexor hallucis brevis; IR, interquartile range; SD, standard deviation. In all analyses, a P-value < .05 with a 95% CI was considered as statistically significant. * Student t test for independent samples was used.

Discussion
To the current knowledge, this research was the first study to analyze and compare the ImageJ software differences in B-mode ultrasound imaging of the intrinsic plantar muscles CSA between hemiparetic and contralateral feet in post-stroke patients. Despite the extrinsic plantar flexor muscles, i.e. the gastrocnemius muscles, were assessed in poststroke survivors [5–7], a lack of research studies of the intrinsic plantar muscles was found. Indeed, this is the first ImageJ software analysis in order to determine the CSA as well as the mean, SD and count of the B-mode ultrasound image pixels.

Regarding the ultrasound measurements from previous studies which evaluated the plantar muscles and fascia in healthy subjects [8–10], our values differed with respect to normalized reference values. Furthermore, the intrinsic plantar muscles were shown to be reduced in patients with pes planus with respect to subjects without this condition [9], as well as in participants with hallux valgus versus subjects without this condition [10].

Currently, new software analyses of the B-mode ultrasound were applied in neural tissue [11,12] and new image pixels analyses were proposed for the assessment of pixel shift in ultrasound images after local temperature changes during the laser interstitial thermotherapy of liver [22]. Nevertheless, future studies about advances in pixel analysis with Image J could help to understand the relationship with neuromuscular alterations and musculoskeletal composition changes.

Limitations

Several limitations could be presented in this study. Despite the incidence of alterations secondary to the intrinsic plantar muscles spasticity was similar between patients with traumatic brain injury and cerebrovascular accident [23], the stroke type associated to hemiparesis was not registered. Furthermore, larger sample sizes and variations of conditions across countries should be considered in future research studies [24].
Conclusion

Image J software differences in B-mode ultrasound imaging of the AbH pixels count were presented between hemiparetic and contralateral feet in poststroke patients.

Conflict of interest and source of funding statement

The authors declare that they have no conflict of interest or source of funding.

Ethical approval

All procedures performed in studies involving human participants were in accordance with the ethical standards of the institutional and/or national research committee and with the 1964 Helsinki declaration and its later amendments or comparable ethical standards. Informed consent was obtained from all individual participants included in the study.

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Abbreviations

AbH, abductor hallucis CSA, cross sectional area

BMI, body mass index

FDB, flexor digitorum brevis

FHB, flexor hallucis brevis

FIM+FAM, Functional Independence Measure and Functional Assessment Measure

MMAS, Modified Modified Ashworth Scale

RUSI, rehabilitative ultrasound imaging

SD, Standard deviation
Figure legends

Figure 1. ImageJ software analysis of the CSA, mean, SD and count of the pixels from the FDB of the hemiplegic and contralateral feet in poststroke patients. Abbreviations: CSA, cross-sectional area; FDB, flexor digitorum brevis; SD, standard deviation.

Figure 2. ImageJ software analysis of the CSA, mean, SD and count of the pixels from the FHB of the hemiplegic and contralateral feet in poststroke patients. Abbreviations: CSA, cross-sectional area; FHB, flexor hallucis brevis; SD, standard deviation.

Figure 3. ImageJ software analysis of the CSA, mean, SD and count of the pixels from the AbH of the hemiplegic and contralateral feet in poststroke patients. Abbreviations: AbH, abductor hallucis; CSA, cross-sectional area; SD, standard deviation.