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A novel cadaveric study of the morphometry of the serratus anterior muscle: one part, two parts, three parts, four?

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Abstract The serratus anterior is portrayed as a homogeneous muscle in textbooks and during functional activities and rehabilitation exercises. It is unclear whether the serratus anterior is composed of subdivisions with distinctive morphology and functions. The purpose of this study was to determine whether the serratus anterior could be subdivided into different structural parts on the basis of its segmental architectural parameters. Eight formalin-embalmed serratus anterior muscles were dissected and the attachments of each fascicle documented. Orientation and size of each fascicle were measured and the physiological cross-sectional area (PCSA) calculated. Three subdivisions of the serratus anterior were identified. A new finding was the discovery of two distinctive fascicles attached to the superior and inferior aspects of rib 2. The rib 2 inferior fascicle had the largest PCSA (mean 1.6 cm²) and attached, with the rib 3 fascicle, along the medial border of the scapula to form the middle division. The rib 2 superior and rib 1 fascicles attached to the superior angle of the scapula (upper division). Fascicles from ribs 4-8/9 attached to the inferior angle of the scapula (lower division). Mean

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fascicle angle relative to a vertical midline reference and PCSA for each division were 29° and 1.3 cm^2 (upper), 90° and 2.2 cm^2 (middle) and 59° and 3.0 cm^2 (lower). This novel study demonstrated the presence of morphologically distinct serratus anterior subdivisions. The results of this study will inform the development of optimal techniques for the assessment, treatment and rehabilitation of this architecturally complex muscle in shoulder and neck pain.

Keywords Anatomy \cdot Cadaver \cdot Neck pain \cdot Serratus anterior \cdot Shoulder pain

Introduction

The accurate assessment of serratus anterior muscle activity is critical to establishing its role in disorders of the shoulder and neck. However, it may be inappropriate to extrapolate the activation of one part of the serratus anterior to the muscle as a whole if it consists of morphologically and functionally distinct subdivisions. Previous electromyographic (EMG) studies have mostly examined the activity of the serratus anterior as a relatively homogeneous muscle during a variety of functional activities and rehabilitation exercises (Alizadehkhaiyat et al. 2015; Huang et al. 2015; Maenhout et al. 2016; San Juan et al. 2016). Typically these studies use electrodes only in one location on the muscle to evaluate the effect of functional activities and exercises on the serratus anterior as one whole muscle. There are no current guidelines or literature to recommend EMG sensor locations to distinguish muscle activity in individual subdivisions of the serratus anterior (SENIAM, Surface Electromyography for the Non-Invasive Assessment of Muscles, http://www.seniam.org). Distinct subdivisions have been demonstrated within

numerous skeletal muscles in cadaveric dissection studies (Gottschalk et al. 1989; Johnson et al. 1994). These anatomical subdivisions are used to inform electrode placement on different parts of the muscle for the accurate determination of muscle activity in each subdivision. No previous studies have used cadaveric dissection to quantify the segmental architectural parameters of the serratus anterior to establish morphologically based subdivisions to inform EMG electrode placement.

The primary role of the serratus anterior is to stabilize the scapula against the thorax and control scapular motion during movements of the shoulder (Lear and Gross 1998; Smith et al. 2003; Castelein et al. 2015). Clinically, dysfunction of the serratus anterior has been implicated in musculoskeletal disorders including shoulder and neck pain (Cools et al. 2014; Castelein et al. 2015). Thus in clinical practice the focus is to use exercise-based therapeutic interventions targeting the serratus anterior in the rehabilitation of patients with shoulder or neck pain (Ebaugh et al. 2005; Witt et al. 2011; Holmgren et al. 2012; Worsley et al. 2013; Cools et al. 2014). However, there is no current literature to recommend effective exercises that target individual subdivisions of the serratus anterior. The delineation of serratus anterior subdivisions will enable the development of guidelines for the accurate placement of surface EMG electrodes and the creation of efficacious serratus anterior strength training and neuromuscular coordination exercises in patients with neck and shoulder pain (Ludewig and Cook 2000; Helgadottir et al. 2011; Sheard et al. 2012; Worsley et al. 2013).

The serratus anterior is typically portrayed as a homogeneous muscle that consists of 9-10 fascicles uniformly arising from ribs 1-8/9 and attaching along the medial border of the scapula (Drake et al. 2010; Moore et al., 2010). In the five primary research studies to examine the morphology of the serratus anterior using cadaveric dissection (Table 1), the muscle is inconsistently subdivided into one, two or three parts. Each subdivision is postulated to have distinctive actions at the scapula (Cuadros et al. 1995; Smith et al. 2003; Ekstrom et al. 2004; Bertelli and Ghizoni 2005; Nasu et al. 2012). No studies have quantified the segmental architectural parameters of the serratus anterior, including fascicle orientation, length and thickness, tendon length and physiological cross-sectional area (PCSA), to establish morphologically based functional subdivisions. Therefore the purpose of this cadaveric study was to determine whether the serratus anterior could be subdivided into different structural parts on the basis of its segmental architectural parameters. The results of this study will be used to inform the placement of SEMG electrodes for the accurate measurement of serratus muscle activity and the development of effective exercises for the management of shoulder and neck pain.

Materials and methods

Eight serratus anterior muscles from formalin-embalmed Caucasian cadavers (one female; three males) aged 69–96 years (mean 84; SD 12 years) were dissected at the Centre for Learning Anatomical Sciences, University of Southampton, UK. Anatomical examination was performed in accordance with the Human Tissue Act, UK (2004), and Anatomy Act, UK (1984), and the study was approved by the institutional ethics committee.

The skin and fascia of the torso and the pectoralis major and minor muscles were removed. The clavicle was disarticulated at the sternoclavicular and acromioclavicular joints and the brachial plexus, axillary vessels and their branches removed to expose the serratus anterior in its entirety (Fig. 1). The fascicular anatomy of the serratus anterior was described and quantified. A fascicle was defined as a bundle of muscle fibers with distinct and identifiable attachments to a rib. The muscle fiber angle of each fascicle was measured at the superior and inferior borders of each rib attachment using a flexible clear plastic goniometer (baseline 360° , measured to the nearest 1.0°). Muscle fiber angle was measured with respect to a vertical midline reference that passed through the suprasternal notch superiorly and the pubic symphysis inferiorly (Fig. 1). Starting from the caudal end of the serratus anterior, each fascicle was systematically detached from its respective rib and followed to its attachment on the scapula from which it was also removed. The sites of attachment were demarcated using colored ink (Cancer Diagnostics Inc., USA), photographed, measured and recorded. The length of the inferior aspect of each rib attachment site and its distance from the vertical midline reference were measured (to the nearest 0.1 cm) using a flexible tape to accommodate the curvature of the thorax (Fig. 2).

After each fascicle had been removed, measurements were made of its size (Fig. 3). If present, tendon lengths at the rib and scapular attachments were measured. After removing the tendinous fibers at each end of the fascicle, the length of its muscle fibers was measured. The thickness of each fascicle was measured at its midpoint and within 10 mm of each attachment end. All length and thickness measures were made from the inferior border of the fascicle using calibrated digital calipers (Absolute Digimatic Calipers CD-6"CX, Mitutoyo Corporation, Japan, measured to 0.01 mm). The volume of the fascicle was measured by submerging it in water, with no splash or loss of water and allowing time for any bubbles to rise to the surface, in a calibrated 100-ml measuring cylinder (VOLAC, Poulten and Graf, Barking, UK) and recording the fluid displacement to the nearest ml after the fascicle had sunk. The PCSA of each fascicle was calculated by

Study	Nasu et al. (2012)	Cuadros et al. (1995)	Smith et al. (2003)	Ekstrom et al. (2004)	Bertelli and Ghizoni (2005)
No. SA (cadavers)	10 (5)	40 (27)	13 (8)	Unspecified (2)	15 (Unspecified)
Gender	4 M; 1F (Japanese)	Unspecified	3F; 5 M	Unspecified	Unspecified
Fixation	Embalmed	Fresh	Unspecified	Unspecified	Fresh
Method(s)	Dissection of whole SA to show structure and innervation of each part	Dissection of lower SA to determine vascular supply	Dissection of upper SA to document attachments	Dissection of whole SA to determine optimal locations for sEMG electrodes	Dissection of whole SA to show structure and innervation of each part
Divisions	Superior part: superior angle and medial border of scapula Middle part: ribs 2–3 to medial border of scapula Inferior part: inferior angle of scapula	Lower part: ribs 6–10 to inferior angle of scapula	Upper: ribs 1–2 (46 %), rib 2 (31 %), rib 1 (15 %) or rib 2–3 (8 %) to superior angle of scapula	Upper part: ribs 1–4 to medial border of scapula Lower part: below rib 4 to inferior angle of scapula	Upper portion: ribs 1–2 to the superior medial border of the scapula Intermediate portion: ribs 2–4 to medial border of scapula Lower portion: ribs 3–8 to inferior angle of scapula
Measures	None	Lower SA flap: Dimensions mean $18.0 \pm 1.9 \times 9.0 \pm 0.8$ (min 12.0 × 8.0; max 21.0×15.0) cm Area mean 164 ± 40 (min 96; max 300) cm ²	Upper SA: Length mean 6.9 ± 1.2 (min 4.8; max 9.0) cm Girth mean 6.1 ± 1.5 cm (min 3.0; max 8.5) cm	None	None
Proposed functions	Superior part: anchor Inferior part: upper rotation of the scapula	Unspecified	Upper SA: compressive load to scapulothoracic joint to anchor/stabilize scapula, anterior tilt of scapula	Upper part: abduction or protraction of the scapula Lower part: upward rotation of the scapula	Upper portion: scapular protraction Lower portion: scapular stabilization

Table 1 Summary of previously reported studies of serratus anterior (SA) morphology

dividing the fascicle volume by its length (Johnson et al. 1994; Bogduk et al. 1998; Phillips et al. 2008).

The serratus anterior was photographed using a digital camera (Nikon Coolpix 5400, Nikon Corporation, Tokyo, Japan) fixed to a tripod (GX-86, OSAWA, Japan), and images were uploaded onto a computer. Descriptive data were presented using mean and standard deviation values for each serratus anterior fascicle.

Fourteen randomly selected fascicles were used for the determination of intra-observer reliability (measurements made 1 week apart by the same observer) and inter-observer reliability (measurements made on the same day by a second observer trained and blinded to the results of the first observer) for each of the measures. Reliability was examined using intra-class correlation coefficients (ICCs). All analyses were performed using SPSS version 17.0 for Windows (SPSS Inc., Chicago, IL, USA) and Microsoft

Office Excel 2003 (Microsoft Corp., Redmond, WA, USA).

Results

Based on the fascicle attachment sites and fiber angles, the serratus anterior was found to consist of three divisions: upper (rib 1 and 2^{S} fascicles attached to the medial and superior borders of the scapula that form the superior angle); middle (rib 2^{I} and 3 fascicles that passed to the medial border); lower (rib 4–8/9 fascicles attached at the inferior angle).

At first sight, the in situ serratus anterior muscle appeared to consist of nine fascicles attaching continuously along the medial border of the scapula. Further dissection revealed up to ten distinct fascicles attached between ribs 1

Fig. 1 Lateral view of the right side of the thorax, clavicle removed and scapula displaced from the thoracic cage, showing the rib 1 (R1) to rib 8 (R8)fascicles of the intact serratus anterior muscle. Two fascicles attach to the superior $(R2^S)$ and inferior $(R2^{I})$ aspect of the second rib. The R1 fascicle is partially obscured by the R2^S fascicle on this view. The measurement of the muscle fiber angle, measured in situ using a flexible goniometer with respect to a vertical midline reference that passed through the suprasternal notch superiorly and the pubic symphysis inferiorly, is depicted for the measurement of the fiber angle at the superior aspect of the right rib 3 fascicle

Fig. 2 Lateral view of the right side of the thorax, clavicle removed and scapula displaced from the thoracic cage, showing the rib 1 (R1) to rib 8 (R8)fascicle rib attachments demarcated by colored ink following removal of the serratus anterior muscle. The rib 2 inferior $(R2^{I})$ and superior $(R2^{S})$ fascicles attached along the inferior border of rib 2 and the superior aspect of rib 2 and first intercostal muscle, respectively. The measurement in situ of the length of each rib attachment and its distance from the vertical midline reference using a flexible tape measure is depicted for the right rib 4 fascicle









10 mm from each end. All measures were made using calibrated digital calipers along the inferior edge of the fascicle

and 9 that arose from the superior angle, inferior angle or medial border of the scapula (Figs. 1, 2, 5). Two distinct fascicles were found to attach to rib 2 and are referred to as the rib 2^{S} (rib 2 superior) and rib 2^{I} (rib 2 inferior) fascicles, based on their rib 2 attachment sites (Figs. 2, 4). In four muscles (three right, one left), the fascicles extended between ribs 1 and 8 with no rib 9 fascicle present. Three small accessory muscle fascicles, with variant muscle fiber angle and/or rib attachment site, were observed (left and right rib 6 and right rib 5). The rib 1 fascicle was absent in one right serratus anterior.

The fibers of the lower division attached to ribs 4–8/9 and passed posteriorly and, to varying degrees, superiorly around the thoracic cage to converge via a common tendon that attached to the inferior angle of the scapula (Fig. 5). Attachment was predominantly to the anterior surface of the inferior angle with the lower three fascicles attaching to the inferior aspect of the inferior angle and extending to the

Fig. 4 Anterolateral view of the thorax, clavicle removed and scapula displaced from the thoracic cage. The rib 3-9 serratus anterior fascicles have been removed to show the attachments of the rib 2 inferior $(R2^{I})$ fascicle, between the medial border of the scapula and the inferior border of rib 2, and the rib 2 superior $(R2^S)$ fascicle, between the superior angle (superior border) of the scapula and the superior aspect of rib 2 and the first intercostal muscle (1st IC), and the rib 1 (R1) fascicle, between the superior angle (medial border) of the scapula and rib 1. a Right side with the R1 fascicle not visible. **b** Left side with the scapula lifted to display the R1 fascicle. **c** Left side with the $R2^{S}$ fascicle removed (attachment sites demarcated) to show the R1 fascicle and the entirety of the R2^I fascicle





Fig. 5 Schematic illustration of serratus anterior fascicle attachments to the right anterior scapula. The rib 2 inferior $(R2^{I})$ fascicle attached along the majority of the medial border of the scapula with the rib 3 (R3) fascicle. The rib 1 and rib 2 superior $(R2^{S})$ fascicles attached to the medial and superior borders of the superior angle, respectively. In half of the muscles dissected, a gap was present between the attachments of the R1 and R2^I fascicles. The rib 4–8/9 fascicles attached to the inferior angle of the scapula

posterior surface. In two muscles, the rib 4 fascicle extended a short distance along the medial border where it was continuous with the rib 3 fascicle. Muscle fibers of the lower four fascicles interdigitated with the external oblique muscle fibers and/or attached to the overlying fascia at their rib attachment. In two muscles, the rib 9 fascicles attached directly to fascia covering the external oblique, rather than the rib.

The middle division predominantly consisted of the rib 2^{I} fascicle, which attached along the curved inferior border of rib 2, ending posteriorly at the anterior aspect of the posterior scalene muscle attachment. Its fibers passed posteriorly to attach along the length of the medial border of the scapula, where it was continuous with the rib 3 fascicle inferiorly and rib 1 fascicle superiorly, with a small gap separating the attachments in approximately half of muscles (Fig. 5).

The rib 2^{s} and rib 1 fascicles of the upper division attached to the superior aspect of rib 2 and rib 1, inferior to the middle scalene muscle attachment, respectively (Fig. 4). In 75 % of muscles, the attachment extended to the fascia of the first intercostal muscle. The fibers of both the rib 1 and rib 2^{s} fascicles passed posteriorly and superiorly to attach to the anterior surface of the superior angle of the scapula (Figs. 4, 5). The rib 1 fascicle attached to the medial border and the rib 2^{S} fascicle to the superior border of the superior angle in all but one of the muscles. The rib 2^{S} fascicle obscured the attachments of the rib 1 and 2^{I} fascicles and was removed first in order to access and measure the angles of the rib 1 fascicle and the superior border of the 2^{I} fascicle (Fig. 4).

The dimensions of each fascicle are presented in Tables 2, 3 and 4. The mean (SD) distance between the vertical midline reference and rib attachment for each division was 11.4 (1.4) cm (upper), 12.4 (1.1) cm (middle) and 14.6 (3.4) cm (lower). The fascicles of the lower division had the longest (medial to lateral) rib attachment 'footprint' and the upper division fascicles the shortest (Table 2). The rib 2^I fascicle was the largest [mean (SD) PCSA 1.6 (0.2) cm^2] (Table 3). If present, the rib 9 fascicle had the smallest mean (SD) PCSA of 0.3 (0.2) cm^2 . The mean (SD) PCSA of the remaining fascicles ranged from $0.5 (0.1) \text{ cm}^2$ to $0.8 (0.3) \text{ cm}^2$ (Table 3). The mean (SD) PCSA for each division of the muscle was 1.3 cm^2 (upper), 2.1 cm^2 (middle) and 3.0 cm^2 (lower). The length of the tendon at each end of the fascicle ranged from no tendinous fibers present to 9.1 mm at the rib attachment and 13.5 mm at the scapular attachment (Table 4). Thickness of the fascicle was greatest at the scapular end compared to the midpoint and rib end (Table 4).

The ICC values for intra-observer (ICC_{3,1}) and interobserver (ICC_{2,1}) reliability were good to excellent for all measurements (Table 5) (Cicchetti 1994). The majority of measurements were excellent (0.85–0.99). The measurements of the fascicle angle and thickness and the size of the rib attachments were the least reliable within raters (0.64–0.68).

Discussion

Distinct subdivisions have been identified within numerous skeletal muscles. This study is the first to confirm the presence of similar subdivisions within the serratus anterior muscle based on characteristic architectural parameters: upper (rib 1 and 2^s fascicles attached to the medial and superior borders of the scapula that form the superior angle), middle (rib 2^{I} and 3 fascicles that passed to the medial border) and lower (rib 4-8/9 fascicles attached at the inferior angle). A novel finding was that the rib 2 fascicle consisted of two parts with the rib 2^I fascicle attaching to the majority of the medial border of the scapula. The presence of these subdivisions requires consideration in the clinical assessment of the serratus anterior muscle using EMG, ultrasound and MRI as well as the development of rehabilitation exercises. The identification of serratus anterior subdivisions with distinctive

Fascicle	No. fascicles	Fiber angle (°) at the	e rib attachment	Rib attachment dimensions (cm)		
		Superior aspect	Inferior aspect	Midline to rib attachment	Rib attachment	
Rib 1	7	27.3 (17.1)	27.7 (16.2)	10.3 (0.7)	2.2 (0.7)	
Rib 2 ^s	8	25.8 (18.7)	29.9 (16.8)	12.3 (1.3)	3.0 (0.6)	
Rib 2 ^I	8	40.9 (17.8)	92.9 (8.4)	12.1 (1.0)	4.6 (1.2)	
Rib 3	8	76.1 (9.0)	86.3 (7.1)	12.7 (1.3)	3.3 (1.0)	
Rib 4	8	70.6 (12.4)	78.4 (9.4)	12.0 (1.0)	4.6 (1.3)	
Rib 5	8	75.4 (8.6)	71.0 (9.6)	11.4 (1.0)	5.3 (1.7)	
Rib 6	8	64.0 (7.6)	62.8 (10.2)	12.9 (0.9)	4.8 (1.1)	
Rib 7	8	47.8 (8.8)	56.1 (6.5)	15.6 (1.5)	4.5 (1.3)	
Rib 8	8	41.6 (10.5)	40.0 (14.3)	18.4 (2.1)	3.6 (1.5)	
Rib 9	4	35.0 (7.7)	27.8 (15.5)	21.7 (0.3)	2.2 (1.5)	

Table 2 Muscle fiber angle measured relative to a vertical midline reference (between the suprasternal notch and pubic symphysis) at the superior and inferior aspect of the rib attachment, distance to the

medial end of the rib attachment from the vertical midline reference and medial-lateral dimension of the fascicle attachment at the rib for each fascicle of the serratus anterior muscle

Mean (standard deviation)

Rib 2^{S} rib 2 superior fascicle, *rib* 2^{I} rib 2 inferior fascicle

Table 3 Length, volume and physiological cross-sectional area (PCSA) (volume divided by length) for each serratus anterior fascicle, identified by their respective rib attachment

Fascicle	No. fascicles	Length (cm)	Volume (ml)	PCSA (cm ²)
Rib 1	7	8.5 (1.5)	4.3 (1.4)	0.5 (0.1)
Rib 2 ^s	8	8.2 (0.9)	6.4 (2.1)	0.8 (0.3)
Rib 2 ^I	8	9.5 (2.2)	15.4 (1.4) ^a	1.6 (0.2) ^a
Rib 3	8	12.6 (1.4)	6.1 (2.5)	0.5 (0.2)
Rib 4	8	14.4 (2.2)	8.5 (2.5)	0.6 (0.1)
Rib 5	8	16.3 (1.8)	8.9 (2.4)	0.6 (0.2)
Rib 6	8	15.9 (2.4)	7.8 (1.4)	0.5 (0.1)
Rib 7	8	16.7 (1.6)	9.1 (3.6)	0.5 (0.2)
Rib 8	8	16.1 (3.1)	8.5 (2.4)	0.5 (0.2)
Rib 9	4	13.2 (2.4)	4.3 (2.6)	0.3 (0.2)

Mean (standard deviation)

Rib 2^{S} rib 2 superior fascicle, *rib* 2^{I} rib 2 inferior fascicle

^a For the calculation of rib 2^{I} PCSA, the mean length of the fascicle along its superior and inferior borders was used [superior border length 7.7 (1.1) and inferior border length 11.3 (1.3) cm]

attachment sites and fascicle angle suggests that these subdivisions do not move and stabilize the scapula in the exact same manner.

The present results support the upper division having a role in controlling and anchoring the superior angle during scapular rotation because of the short thick rib 1 and 2^{s} fascicles attached to both borders of the superior angle of the scapula and orientated closer to the vertical compared to middle and lower division fascicles. Previous authors have proposed that the serratus anterior anchors the scapula

by pulling the superior angle to the ribs to enable rotation of the scapula by influencing the main axis of scapular rotation (Gregg et al. 1979; Hamada et al. 2008, Martin and Fish 2008; Nasu et al. 2012). Interestingly, we observed these fibers passing anteriorly from the scapula to the ribs suggesting these fibers do have a role in anchoring the superior angle to the ribs and controlling movements of the scapula by influencing the axis of rotation, which changes during elevation. We propose that the upper division, because of its attachment to both the superior and medial borders that form the superior angle and the inferior orientation of the fibers as they pass to the ribs, could play a role in controlling external rotation of the scapula by anchoring the superior angle. Relative upward rotation of the scapula is maintained by keeping the acromion above the superior angle. The authors also suggest that the upper division, by anchoring the superior angle to ribs 1 and 2 (positioned inferior to the superior angle), helps to maintain optimum scapular orientation and relative scapula upward rotation, by keeping the superior angle inferior to the acromion. The longer tendon of the rib 2^S fascicle at the scapular end could contribute to directing the forces required to anchor the superior angle.

The rib 2^I fibers passed virtually horizontally to their rib attachment, and despite being the thinnest fascicle (Table 4), we discovered the fascicle had the largest PCSA. We suggest this supports the role of the middle division for producing external rotation (and controlling internal rotation) of the scapula at the acromioclavicular joint and then protraction of the clavicle at the sternoclavicular joint, as described by Ludewig and Reynolds (2009). However, before protraction can occur, the line of action of the

Fascicle	Fascicle thickness (mm)				Tendon length (mm)			
	No. fascicles	Scapula end	Midpoint	Rib end	No. fascicles	Scapula end	No. fascicles	Rib end
Rib 1	7	3.1 (1.3)	2.5 (2.1)	2.0 (0.8)	1	1.8	1	3.5
Rib 2 ^s	8	3.1 (1.2)	3.4 (1.7)	2.6 (1.0)	2	12.0 (14.5)	4	3.2 (1.7)
Rib 2 ^I	8	2.4 (0.6)	1.9 (1.3)	1.3 (0.5)	4	4.8 (0.6)	4	3.9 (0.8)
Rib 3	8	3.4 (1.2)	1.9 (0.8)	1.5 (0.8)	7	6.0 (4.1)	1	9.1
Rib 4	8	4.2 (1.6)	2.0 (0.6)	0.9 (0.3)	7	7.0 (4.4)	3	2.4 (1.1)
Rib 5	8	4.2 (1.4)	3.8 (1.5)	1.1 (0.4)	6	5.0 (2.2)	3	6.4 (3.9)
Rib 6	8	4.0 (2.4)	3.8 (0.8)	1.4 (0.3)	6	5.5 (3.0)	4	3.3 (1.1)
Rib 7	8	4.9 (2.1)	4.0 (1.3)	1.5 (0.3)	7	4.9 (2.1)	1	2.4
Rib 8	8	3.5 (1.1)	4.0 (1.0)	1.8 (0.7)	8	5.5 (2.4)	1	8.4
Rib 9	4	2.7 (1.5)	2.5 (0.9)	1.3 (0.4)	2	13.5 (2.5)	1	6.1

Table 4 Thickness of the muscle fibers within each fascicle, measured along the inferior border at the mid-point of each fascicle and 10 mm from both the lateral scapular end and the medial rib end following removal of the tendon

The length of the tendon, when present, was measured along the inferior border at the lateral scapular end and the medial rib end of each fascicle. Mean (standard deviation)

Rib 2^{s} rib 2 superior fascicle, *rib* 2^{I} rib 2 inferior fascicle

 Table 5 Reliability for each measure of serratus anterior fascicle morphometry

Fascicle measurement	Intra-observer (ICC _{3,1})	Inter-observer (ICC _{2,1})
Angle	0.64	0.94
Length	0.96	0.94
Thickness	0.85	0.74
Volume	0.96	0.85
Rib attachment to midline	0.99	0.97
Rib attachment	0.68	0.70

Ratings for clinical significance of ICC values (Cicchetti 1994): <0.40 poor; 0.40–0.59 fair; 0.60–0.74 good; 0.75–1.00 excellent *ICC* intraclass correlation coefficient

serratus anterior will first pull the medial border and inferior angle of the scapula towards the chest wall, creating external rotation of the scapula. This external rotation of the scapula will stabilize the scapula as protraction of the clavicle at the sternoclavicular joint occurs. The rib 2^I fascicle attachment along the medial border of the scapula suggests this fascicle has a role in controlling scapular winging, which is a commonly observed problem in shoulder pain and dysfunction (Ludewig and Reynolds 2009). Because of the large PCSA of this fascicle, consideration needs to be given to its force capabilities and the influence this could have on controlling scapular winging. Calculation of the force capacity of the serratus anterior fascicles and subdivisions would help support this proposition.

The lower division fascicles were of approximately equal dimensions and PCSA. At the scapula, the muscle attachment is concentrated at the inferior angle and projects anteriorly and inferiorly to attach to ribs 8/9-4 at an angle increasing from 28° to 78°. The fibers will pull the inferior angle laterally around the chest wall (away from the midline), and because the axis of rotation is slightly inferior to the spine of the scapula, this supports a primary role of upward rotation of the scapula. The increasing angle of the fibers to rib 4 suggests the fiber orientation changes in relation to the rib position to optimize the pull of the inferior angle laterally. The role of upward rotation is confirmed by EMG studies of the lower division where exercises that create upward rotation of the scapula produce greater EMG activity compared to straight scapular protraction exercises (Moseley et al. 1992; Ekstrom et al. 2004). In participants with shoulder impingement, Worsley et al. (2013) reported delayed onset and early termination of serratus anterior activity using surface EMG, and less posterior tilt and upward rotation with three-dimensional kinematic analysis. EMG was recorded from the lower serratus anterior division as described by Ludewig and Cook (2000). The results of the present study are in agreement with previous authors that have suggested the lower division produces upward rotation (Ludewig et al. 2004; Roren et al. 2013) and controls downward rotation (Worsley et al. 2013). Activity of the serratus anterior has been noted in scapular posterior tilt exercises (Ha et al. 2012), and posterior tilt has been noted in people with long thoracic nerve lesions (Roren et al. 2013) supporting its role in scapular posterior tilt. The authors suggest posterior tilt is produced by the inferior laterally directed fibers of the lower division and the middle division is likely to contribute to controlling anterior tilt too by keeping the medial border close to the thoracic spine. Further research is needed to explore the degree of muscle activity for each serratus anterior subdivision during a variety of rehabilitation exercises. Greater delineation of which exercises optimally recruit each subdivision is of interest to clinicians as this may inform the development of more effective treatment and exercise programs.

The optimal EMG electrode placement location for the serratus anterior is not known. Clearly the serratus anterior has distinctive divisions that require consideration when evaluating this muscle and developing effective clinical tests and training strategies. The results of the present study provide the anatomical basis for the development of new protocols for surface EMG electrode placement to ensure that electrodes are positioned parallel to muscle fibers and on muscle rather than tendon (Hermens et al. 2000). Furthermore, the detailed morphometry reported in the present study can be used to establish standardized sites and valid and reliable methods of applying ultrasound and MR imaging for the measurement of muscle geometric parameters and functions for each serratus anterior subdivision (Talbott and Witt 2013). To measure muscle activity in the upper division, it is suggested that electrodes be placed on the more superficial rib 2^s fascicle rather than the rib 1 fascicle, given that they have equivalent fiber angle and PCSA. The inferior part of the rib 2^I fascicle, where the muscle fibers are oriented at 90° to the midline, is recommended for electrode placement on the middle division. Not only is the rib 2^I fascicle the broadest and most distinctive in the middle division, it also represents the majority of the middle division PCSA. The rib 6 fascicle is recommended for electrode placement on the lower division as it is both accessible and characteristic of this division. The placement of surface electrodes on both the rib 2^S and 2^I fascicles is likely to be challenging because of the overlying scapula and presence of nearby muscles such as the pectoralis major and minor. Future research using ultrasound and diffusion tension magnetic resonance imaging in living subjects is recommended to refine these cadaveric-based recommendations and enhance the accurate acquisition of serratus anterior muscle activity from each subdivision.

The present study has a number of limitations. First, the cohort studied was small, but as an exploratory study the results are promising, and, in our view, because of the clinical relevance of the serratus anterior in shoulder and neck pain, a larger study is warranted. A larger sample size would enable investigation of the consistency of the serratus anterior morphology and prevalence of the variations documented in this and previous studies (Eisler 1912; Cuadros et al. 1995; Smith et al. 2003). Second, the quantification of

muscle volume, using the Archimedes' principle of fluid displacement, can be affected by the extent of muscle hydration (Ward and Lieber 2005) and the presence of air bubbles. Attempts were made to minimize these factors by the use of 5 % formaldehyde solution for embalming and allowing time for any bubbles to rise to the surface. In addition, while great care was taken to define the muscle attachment and remove it directly from its attachment site, it is possible that some periosteum may have been removed in this process, creating an artifact. Finally, the challenge of measuring the serratus anterior fiber angle is exemplified by the lower intra-observer reliability value compared to other measurements (Table 5). Different measurement methods have been employed to measure the muscle fiber angle but no gold standard exists (De Foa et al. 1989; Johnson et al. 1994; Ackland et al. 2008). Serial dissection combined with digitization and three-dimensional modeling in cadavers and diffusion tensor imaging in vivo are promising new methods for the visualization and quantification of muscle architecture throughout the entire muscle volume (Lee et al. 2015). A future study powered for the determination of anatomical variants and with some methodological improvements, such as digital measurement of architectural parameters, is justified.

The findings from this novel study of the fascicular architectural morphometry of the serratus anterior muscle suggest that the muscle consists of three distinctive subdivisions (upper, middle and lower). Although these preliminary results need confirmation with a larger study, they will inform accurate location of electrodes during SEMG assessment of the serratus anterior and the functional relevance of the subdivisions. The findings have clinical implications for the development of optimal techniques for the assessment, management and rehabilitation of this architecturally complex muscle.

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Compliance with ethical standards

Conflict of interest Sarah Mottram is Director of Movement Performance Solutions, Ltd., and educates and trains sports, health and fitness professionals to better understand, prevent and manage musculoskeletal injury and pain that can impair movement and compromise performance in their patients, players and clients. The remaining authors have no conflict of interest to declare. No financial support or equities were provided by Movement Performance Solutions or other sources.

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