The aim of this study was to investigate the functions of the six subregions of the supraspinatus muscle (SSP) determined by Kim et al. in Clin Anat 2007;20:648–655, using real-time tissue elastography (RTE). Twelve young male volunteers participated. The muscular hardness of the SSP was measured at rest and with contraction of the MMT3 in internal, neutral and external rotations. The SSP was functionally divided into five groups on the basis of the RTE results. These functional areas were roughly classified into three property groups: the anterior-superficial, anterior-middle, and anterior-deep subregions, which produce contractile force for abduction; the posterior-deep subregion, which produces contractile force for external rotation; and the posterior-superficial and posterior-middle subregions, which maintain tension. RTE was appropriate for measuring the functions of these muscular subregions. Clin. Anat. 30:347–351, 2017.
RTE could demonstrate the recruitment of the muscular fasciculus under various contraction conditions using color mapping (Fig. 1).

**Equipment**

A diagnostic ultrasound system (Noblus; Hitachi-Aloka Medical Japan, Tokyo, Japan) with a linear array probe (L-64; Hitachi-Aloka Medical Japan, Tokyo, Japan) was used. An acoustic coupler (EZU-TECPL1; Hitachi-Aloka Medical Japan, Tokyo, Japan), with an elastic modulus of $22.6 \pm 2.2$ kPa was placed on the probe with a plastic attachment (EZU-TEATC1; Hitachi-Aloka Medical Japan, Tokyo, Japan).

We oriented the cross-sectional ultrasound B-mode sonograms by placing the probe 2 cm medial to the acromion and obtained the RTE images by applying cyclic manual compression with it. The result was a color-coded map with harder tissue represented in blue and softer tissue in red.

**RTE Assessment of the SSP**

Using Kim’s six subregions, namely anterior-superficial (AS), anterior-middle (AM), anterior-deep (AD), posterior-superficial (PS), posterior-middle (PM), and posterior-deep (PD), we located seven ranges of interest (ROIs) on the cross-sectional RTE images. Because the AM has an intramuscular tendon that is hard relative to muscular tissue, we measured the elasticities of the anterior and posterior muscular parts of the AM subregion (AM-a and AM-p) excluding the tendon. We set a square ROI A on an acoustic coupler and a circular ROI B on target subregions and then calculated the strain ratio ($SR = B/A$). The average SR value of three randomly selected images was adopted as “the SR value” for each task. We defined the SR value at rest and during contraction as “the SR value at rest” and “the SR value at contraction”, respectively. The SR value at rest indicates the muscle hardness without contraction and is affected by muscle length; that is, muscles become harder when they lengthen and softer when they shorten. The SR value at contraction is a sum of the muscle’s hardness at rest and the hardness resulting from contraction. We defined the difference between the SR values at rest and during contraction as the activity value.

**Statistics**

R version 2.8.1 was used for statistical analyses. One-way repeated ANOVA followed by Shaffar’s modified sequentially rejective Bonferroni procedure and Friedman’s test with Wilcoxon’s post hoc test were used to compare the SR values among three rotational positions. Friedman’s test with Wilcoxon’s post hoc test was used to compare the SR values among subregions. Statistical significance was set at 0.05.

**RESULTS**

At rest, the AS, AM-a, AM-p, and AD showed a mountain-shaped elastograph with neutral rotation at the top. The SR value for the neutral rotation task was significantly higher than those for the external and internal rotation tasks in all subregions except the AM-p, including the AR. In the AM-p, there was no difference between the SR values for the neutral and external rotation tasks. The AD had smaller SR values than the AS, AM-a, and AM-p. The SR values for the three rotational positions were not significantly different for the PS and PM, while the values for external and neutral rotation were higher than that for internal rotation in the PD. The SR value for external rotation was the highest but did not differ significantly from the neutral rotation value in the PD. During contraction, there was no significant difference among the three rotational positions for any subregion except the AS, where the SR value for external rotation was significantly higher than for neutral and internal rotation, though the difference was small (Fig. 2).
We compared the highest SR values at rest and contraction (blue line) of the subregions in three rotational positions were plotted. *P < 0.05, **P < 0.01, and ***P < 0.001. At rest, AS, AM-a, AM-p, and AD had a mountain shape with neutral rotation at the top, while PS and PM had a flat pattern. The values for the external and neutral rotations were higher than those for internal rotation for PD. At contraction, there was no significant difference among the three rotational positions for any subregion except AS. The value for external rotation was significantly higher than those for the neutral and internal rotations, but the difference was small. [Color figure can be viewed at wileyonlinelibrary.com]

Therefore, the SSP comprises five distinct subsections: the AS, AM, AD, (PS and PM), and PD.

**Fig. 2.** The SR values at rest (red line) and at contraction (blue line) of the subregions in three rotational positions were plotted. *P < 0.05, **P < 0.01, and ***P < 0.001. At rest, AS, AM-a, AM-p, and AD had a mountain shape with neutral rotation at the top, while PS and PM had a flat pattern. The values for the external and neutral rotations were higher than those for internal rotation for PD. At contraction, there was no significant difference among the three rotational positions for any subregion except AS. The value for external rotation was significantly higher than those for the neutral and internal rotations, but the difference was small. [Color figure can be viewed at wileyonlinelibrary.com]

| Subregions       | AS | AM-a | AM-p | AD  | PS | PM |
|------------------|----|------|------|-----|----|----|
| AM-a             | 0.053 |      |      |     |    |    |
| AM-p             | 0.053 | 1.000 |      |     |    |    |
| AD               | 0.053 | 0.136 | 0.065 |     |    |    |
| PS               | 0.053 | 0.053 | 0.053 | 0.053 |    |    |
| PM               | 0.053 | 0.067 | 0.053 | 0.067 | 0.053 | 0.053 |
| PD               | 0.053 |      |      |     |    |    |

There was no difference between AM-a and AM-p or between PS and PM.

**TABLE 2. Comparison of the Largest Activity Values of the Six Subregions**

| Subregions       | AS | AM-a | AM-p | AD  | PS | PM |
|------------------|----|------|------|-----|----|----|
| AM-a             | 0.081 |      |      |     |    |    |
| AM-p             | 0.053 | 1.000 |      |     |    |    |
| AD               | 0.053 | 0.196 | 0.081 |     |    |    |
| PS               | 0.053 | 0.053 | 0.053 | 0.053 |    |    |
| PM               | 0.053 | 0.053 | 0.053 | 0.053 | 0.419 |    |
| PD               | 0.053 | 0.061 | 0.053 | 0.061 | 0.061 | 0.081 |

There was no difference between AM-a and AM-p.
DISCUSSION

The muscular hardness at rest changed with shoulder rotation. This phenomenon reflected changes in the footprint orientation. When the long axis of the subregion coincided with the direction of the axis of the entire muscle, which changed with shoulder rotation, the subregion was shortest. The highest SR value at rest indicated the lowest muscular length, and lower SR values indicated a longer muscle. Gates et al. clearly demonstrated how the AR and PR changed their footprints and their long axis directions with shoulder rotation (Gates et al., 2010). Kim et al. (2007) in a morphological study, divided the SSP into six subregions and revealed that these subregions had an appropriate length and direction relative to the long axis. They also found that the subregions of the AR were longer and larger than those included in the PR (Kim et al., 2007).

In this study, the SR values at rest in the AS, AM, and AD had a mountain shape and were larger in the three rotational positions, while the SR values of the PS and PM did not differ with rotational position. Additionally, the PS and PM had lower SR values than the AS, AM and AD. These results suggest that the muscles in the AS, AM, and AD were longer than those in the PS and PM. The differences in SR values for internal and external rotation in the subregions composing the AR reflected differences in the long axes of these subregions.

The SR value at rest was highest for the AS and rather small for the AD. In contrast, the SR values at rest for the PS and PM were smaller than that for the PD. Because the abduction position differed in these subregions, it was difficult to compare the results for this position directly. In a shear wave elastography study, Itoigawa reported that the deep region of the AR and the superficial region of the PR were harder than the superficial region of the AR and the deep region of the PR (Itoigawa et al., 2015). It was likely that the subregions had appropriate hardness at rest. Therefore, in this study, each subregion showed a characteristic pattern with rotation.

During contraction, the SR values of the six subregions were concentrated around the same value. The subregions might work together and finally act as one muscle during contraction.

The changes in activity value resulted from muscular contraction, reflecting the contractile properties of the muscles. Larger and smaller activity values indicate the ability to exert larger and smaller contractile forces, respectively. The large activity values noted for all three rotational positions for the AS, AM, and AD indicate that these subregions functioned powerfully in all rotational positions. The moderate values observed for external and neutral rotation and the lower values for internal rotation in the PD show that the PD functioned forcefully during external and neutral rotation but did not generate internal rotation. The small activity values observed for the three rotational positions of the PS and PM indicated that these subregions did not induce motion but maintained tension in every position. Therefore, we concluded that the AS, AM, and AD exert a large contractile force and act as movers, while the PS and PM retain muscular tension with limited contractile properties and act as stabilizers.

Apart from the results for the PD, these findings are consistent with previous studies. Rho et al. reported that the AR was responsible for contractile force and that the PR could not generate large contractile loads (Rho et al., 2000). Kim et al. reported that the AR was more likely to be responsible for force production and that the PR might maintain tension on the tendon (Kim et al., 2010).

Gates and colleagues reported that the AR induced either external or internal rotation while the PR only contributed to external rotation (Gates et al., 2010). Ackland and Pandy (2011) showed that the PR was a more effective external rotator during abduction. In the present study, the subregions included in the AR functioned powerfully in all rotational positions, but the PS and PM did not induce motion. Only the PD functioned during the external and neutral rotations. The PD was an external rotator during 90° shoulder abduction. The subregions had characteristic functions resulting from differences in the volume and direction of their long axes; those included in the AR functioned powerfully in all rotational positions, the PD acted somewhat forcefully in external and neutral rotations, and the PS and PM only maintained tension in every position.

There were limitations to the present study. First, measurements were only obtained with the shoulder at 90° abduction because this position was selected for performing the MMT3 task. Future investigation using another glenohumeral joint position is needed.

In conclusion, the SSP was functionally divided into five groups using RTE. These regions were roughly classified according to three properties and characterized as follows: the AS, AM, and AD subregions, which produce contractile force for abduction; the PD subregion, which produces contractile force for external rotation; and the PS and PM subregions, which maintain tension. RTE was a suitable method for measuring the functions of the muscular subregions.

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