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● *Original Contribution*

THE SKIN ACTS TO MAINTAIN MUSCLE SHEAR MODULUS

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Abstract—It is not clear how the tissues covering the skeletal muscles affect the muscles' mechanical properties. The main purpose of this study was to examine changes in muscle shear modulus as a representative mechanical property of muscle with and without the covering tissues of skin and epimysium (fascia). Shear modulus of the medial gastrocnemius (MG) muscle was determined using ultrasound shear-wave elastography in the Thiel's embalmed cadavers under three different conditions: original (intact cadavers), removal of the skin on the MG and subsequent removal of the epimysium. Muscle shear modulus significantly decreased by 50% after removal of the skin, whereas no additional changes in shear modulus were observed after subsequent removal of the epimysium. This study suggests that the skin is a main contributor for maintaining the muscle mechanical properties among tissues covering the skeletal muscle. (E-mail: y-yoshi@nifs-k.ac.jp) © 2015 World Federation for Ultrasound in Medicine & Biology.

Key Words: **Shear-wave elastography, Muscle stiffness, Cadaver, Epimysium, Muscle mechanical properties.**

INTRODUCTION

In mammal, the skin is a layer of tissue that covers the body surface area. As the outermost layer of protection, the skin is quite versatile in that it protects the body from a wide range of threats, *e.g.*, biochemical, biologic, immunologic, physical, mechanical and thermal damage (Elias 2005). From a mechanical point of view, the skin has a viscoelasticity that (i) helps hold together the internal tissues and organs (Diridollou et al. 1998; Grahame and Holt 1969) and (ii) allows the skin to act as a shock absorber, protecting the internal tissues from external impulses (Elias 2005). However, despite the skin being in close proximity and partly covering the skeletal muscles, the effect of the skin on the underlying muscle mechanical properties is not well understood.

In humans, the skin is expected to oscillate during muscle contractions or limb movements. Even during static (isometric) contractions, for example, the activations of motor units induce oscillations in the skin on the muscle caused by muscle deformation (Yoshitake et al. 2002, 2008). Furthermore, during dynamic contractions, skin oscillations can occur from impact with the floor during landing (Kraemer et al. 1998) or inertia at onset of limb movements. Interestingly, these oscillations have been shown to be reduced when wearing elastic compression garments at low pressure (at around 20 mmHg), accompanied with an enhancement in jump height during repetitive jumping (Doan et al. 2003; Kraemer et al. 1998). These observations suggest that the tissues covering the muscle play an important role in avoiding unwanted muscle deformation. Interestingly, it has been shown that de-loading tape applied to the muscle belly reduced stress in the underlying muscle regions (Hug et al. 2014). Taken together, this implies that the tissues covering the skeletal muscle, such as skin or epimysium (fascia), must contribute to the underlying muscle mechanical properties. In other words, the muscle mechanical properties, such as muscle stress, would degrade if the tissues covering the skeletal muscle were removed. No studies, however, have yet examined how the tissues covering the skeletal muscle act to maintain the muscle mechanical properties. This may partly be due to difficulties in accurately measuring the muscle

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mechanical properties. In addition, a proper investigation into the mechanical interaction between skin or epimysium and muscle requires the removal of such tissues covering the skeletal muscles in humans *in vivo*.

On the other hand, recent technological developments for assessing such muscle mechanical characteristics—in particular, so-called ultrasound shear-wave elastography—enable us to non-invasively measure the muscle shear modulus (muscle stiffness) with simple operations and high reproducibility (Nordez and Hug 2010; Shinohara et al. 2010; Yoshitake et al. 2014). As a representative mechanical property of muscle, muscle shear modulus has been extensively measured under various experimental conditions, such as at rest (Shinohara et al. 2010; Yoshitake et al. 2014), during muscle contraction (Nordez and Hug 2010; Shinohara et al. 2010; Yoshitake et al. 2014) and passive stretching (Eby et al. 2015; Hug et al. 2013; Lacourpaille et al. 2013; Maïsetti et al. 2012; Taniguchi et al. 2015). Therefore, using shear-wave elastography, the present study examined changes in muscle shear modulus with and without the covering layers of skin and epimysium in humans. To this end, we examined skeletal muscle obtained from Thiel's embalmed cadavers (Benkhadra et al. 2009). Thiel's method is considered to be a valuable technique in clinical and surgical practice because it preserves the flexibility observed in living humans (Benkhadra et al. 2009, 2011; Holzle et al. 2012; Thiel 2002). Through use of the Thiel's embalmed cadavers, we will gain better insight into the direct effect of the skin and epimysium on the muscle shear modulus.

In the experiment mentioned above, if the removal of the muscle-covering tissues significantly affects the muscle shear modulus, one might doubt that the observed changes in shear modulus are caused primarily by changes in muscle architecture, most notably through pennation angle—in other words, angle between the probe (direction of transverse axis of the imaging plane) and longitudinal direction of the fascicle—due to deformation of the muscle after removal of the covering tissues. Therefore, we also attempted to examine the effect of the angle between the probe and longitudinal direction of the fascicle on shear modulus measurements.

The main purpose of this study was to examine changes in muscle shear modulus with and without the layers of skin and epimysium. We hypothesized that muscle shear modulus would be lower in muscle without the surrounding layers of tissue.

METHODS

Six legs from four human cadavers (three males and one female; mean age at death, 89.3 ± 7.5 y; reported antemortem height, 154.3 ± 6.0 cm; reported antemortem weight, 46.3 ± 5.1 kg, mean ± standard deviation) were fixed using Thiel's method (Thiel 1992a, 1992b, 2002). The causes of death were senile decay and cerebral infarction. This experimental protocol was approved by the Ethics Committees of the School of Medicine, Sapporo Medical University.

Experimental setup

The dissections on the cadavers in this study (removal of the skin or epimysium and cutting the muscle) were performed by an anatomist of the School of Medicine, Sapporo Medical University with 10 y of experience. All cadavers were laid facedown on a specially designed bed with knees angled at 170 degrees (180 degrees: full extension) throughout the experiments. Muscle shear modulus by ultrasound shear-wave elastography was measured by another experimenter who was familiar with experiments using ultrasound shear-wave elastography (Miyamoto et al. 2015).

Ultrasound shear-wave elastography

Shear modulus of the medial head of gastrocnemii (MG) was obtained by using an ultrasonic scanner for musculoskeletal testing with shear-wave elastography (AixPlorer Ver. 6.3, Supersonic Image, Aix-en-Provence, France). Shear modulus (KPa) values were obtained from the shear wave propagation velocity in the direction of the longitudinal axis of a probe (4–15 MHz, SL15-4, linear-array, 50-mm wide, Supersonic Imagine, Aix-en-Provence, France) using built-in software. The spatial average of shear modulus in the selected circular area was measured using software (Q-BoxTM) installed in the ultrasound system (Taniguchi et al. 2015; Yoshitake et al. 2014). Care was taken not to press or deform the muscle with the probe during measurements. A single examiner placed the probe on the muscle surface and conducted all measurements.

Experiment 1

To examine the effect of the skin on muscle shear modulus, muscle shear modulus was measured using the above-mentioned ultrasound scanner before and after removal of the skin and subsequent removal of the epimysium. The angle of the ankle joint was kept constant at around 10 degrees plantarflexion from the anatomic position (defined as 0°, with larger values signifying plantarflexion) by placing a cuboid expanded polystyrene under the ankle throughout experiment 1.

The ultrasound probe was placed over the muscle belly of MG. The probe was aligned with the direction of the fascicle to measure muscle shear modulus so that