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Muscle, tendon, and somatotropin responses to the restriction of muscle blood flow induced by KAATSU-walk training

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Summary

Objective: **The efficacy of KAATSU training has been demonstrated in human athletes, both as a therapeutic method as well as a training aid. The purpose of this study was to investigate the effects of slow walk training combined with restriction of muscle blood flow (KAATSU) on muscle and tendon size.**

Methods: **Six healthy, unfit Standardbred mares performed walking (240 m/min for 10 min and then 5 min recovery) with KAATSU, and 6 mares performed walking without KAATSU. A specially designed elastic cuff[1] was placed at the most proximal position of the forelegs and inflated to a pressure of 200–230 mmHg throughout the walking and recovery sessions. The training was conducted once a day, 6 days/week for 2 weeks. Skeletal muscle thickness and tendon thickness were measured using B-mode ultrasound at baseline and after 2 weeks of training. Venous blood samples were obtained before the first acute exercise and 5, 15 and 60 min afterwards. Serum somatotropin concentration was determined using a commercially available equine-specific ELISA kit.**

Results: **The acute increase in plasma somatotropin was 40% greater (P<0.05) in the KAATSU-walk group than in the Control-walk group 5 min after exercise and remained elevated (P<0.05) at 15 and 60 min post exercise compared with the Control-walk group. After 2 weeks of training, muscle thickness increased (P<0.05) 3.5% in the KAATSU-walk group but did not change in the Control-walk group (0.7%). Tendon thickness did not change (P>0.05) in either group.**

Conclusions: **These data demonstrate that KAATSU training can induce muscle hypertrophy in horses and suggest that KAATSU training may provide significant therapeutic/ rehabilitative value in horses, as has been shown in man.**

Introduction

It is well established that the magnitude of the acquired training adaptation is proportional to the training stimulus. In man, to produce significant muscle hypertrophy and strength gains, a training intensity >65% of 1-repetition maximum (1-RM) is required (McDonagh and Davies 1984; Campos *et al.* 2002). Low-intensity resistance training (LIT; ~20-50% 1-RM) rarely produces substantial muscle hypertrophy or strength gains (Kraemer and Ratamess 2004) while high-intensity resistance training (HIT; >65% of 1-RM) increases the risk of injury, especially for frail and elderly populations (McDougall *et al.* 1985; Haykowsky *et al.* 1996).

Low-intensity resistance exercise, when combined with restriction of venous blood flow from the working muscle (KAATSU), has been consistently shown to increase muscular size and strength (Shinohara *et al.* 1998; Takarada *et al.* 2000b; Burgomaster *et al.* 2003; Abe *et al.* 2005). KAATSU, combined with LIT, produces similar increases in muscle cross-sectional area (CSA) as HIT and 3 times higher growth hormone (somatotropin) secretion in man (Takarada *et al.* 2000a). In fact, a training stimulus as low as walking, when combined with KAATSU, has been shown to produce substantial increases in muscle size and strength in man (Kearns *et al.* 2005).

Orthopaedic problems are the primary cause of poor race performance in horses; they also prevent the equine athlete from training. The use of KAATSU training may be beneficial to horses, not only as a preventive measure, but also when they cannot tolerate high-intensity exercise, such as during the recuperation period following injury. Previously, we demonstrated that acute KAATSU results in a significant but transient increase in the thickness of the *ulnaris lateralis*/flexor *digitorum profundus* (UL-FDP) and extensor *digitorum communis* (EDC) muscles in horses (Abe *et al.* 2004). That study also demonstrated the safety of the KAATSU technique in horses and led to the hypothesis that KAATSU plus walking would result in gains in muscle size similar to the gains seen in man. However, there are no data on the effects of chronic KAATSU combined with light exercise in horses. Therefore, the purpose of the present study was to test the hypothesis that slow walk training with KAATSU will result in greater increases in muscle and tendon size than walk training by itself.

Materials and methods

Animals

Twelve healthy, unfit Standardbred mares (mean ± s.d., age 12 ± 4 years, body mass 518 ± 55 kg) were used for the study. The mares were randomly divided into 2 training groups: walk-

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training with restricted leg muscle venous blood flow (KAATSU-walk, n = 6), and walk-training without restricted leg muscle blood flow (Control-walk, n = 6). All methods and procedures were conducted with the prior approval of the Rutgers University Institutional Review Board for the Care and Use of Animals.

General experimental design and training protocol

Before the experiment, the horses were trained to wear a pressure cuff belt[1] at the most proximal position of the foreleg. During the acclimatisation period, the external pressure of the cuff (130–160 mmHg) was selected with regard to their resting blood pressure as described previously (Abe *et al.* 2004). During the acclimatision period, no sign of discomfort or pain was observed in the horses. Muscle blood flow below the pressure cuff has been measured previously in horses using pulsed-wave doppler on the radialis artery during KAATSU. These data demonstrated that blood flow was restricted but not completely occluded (Abe *et al.* 2005a).

The horses in both the KAATSU-walk and Control-walk groups performed 2 weeks of walk training. Training was conducted once a day, 6 days/week for 2 weeks. Following limb girth measurements, the horses performed walking (240 m/min for 10 min) on a motor-driven treadmill and then 5 min standing for recovery. The walking speed and duration remained constant throughout the training period.

Horses in the KAATSU-walk group wore the pressure belt on both forelegs during training. Prior to the KAATSU-walk training, the horses were acclimatised to wearing the pressure cuffs whilst standing on the treadmill, and the belt air pressure was repeatedly set (60 sec) and then released (10 sec) from the initial (140 mmHg) to final (180 mmHg) pressure. On the first day of the training, the final belt pressure (training pressure) was 180 mmHg. The pressure was increased by 10 mmHg each day until a final belt pressure of 230 mmHg was reached. A restriction pressure of 180–230 mmHg was selected for the restriction stimulus because horses adapted to the restriction stimulus during the training. The pressure was based on a review of the data in man (Abe *et al.* 2005a). Foreleg muscle blood flow was restricted for a total time of about 15 min (10 min walking and 5 min recovery) for each horse. The Control-walk group performed the same exercises at the same speed but without KAATSU.

Blood sampling and hormonal assay

On the first day of the training, venous blood samples were obtained prior to the start of training exercise, and during recovery, immediately after exercise and again at 15 and 60 min after exercise. Resting blood samples were also taken at midpoint and 2 days after the last training session. Serum somatotropin concentration was determined using a commercially available equine ELISA kit[2].

Skeletal muscle and tendon size measurements

Skeletal muscle thickness (EDC) and tendon thickness (superficial/deep digital flexor tendon, SDFT) were measured using B-mode ultrasound prior to the training and 2 days after the last training session. The measurement site for the EDCmuscle was midway between the olecranon and the accessory carpal bone. Measurements were made using a 5 MHz scanning head coated with vegetable oil and placed perpendicular to the tissue interface. The subcutaneous adipose tissue-muscle interface and intermuscular interface were identified from the ultrasound image. The distance from the adipose tissue-muscle interface to the intermuscular interface was accepted as the muscle thickness

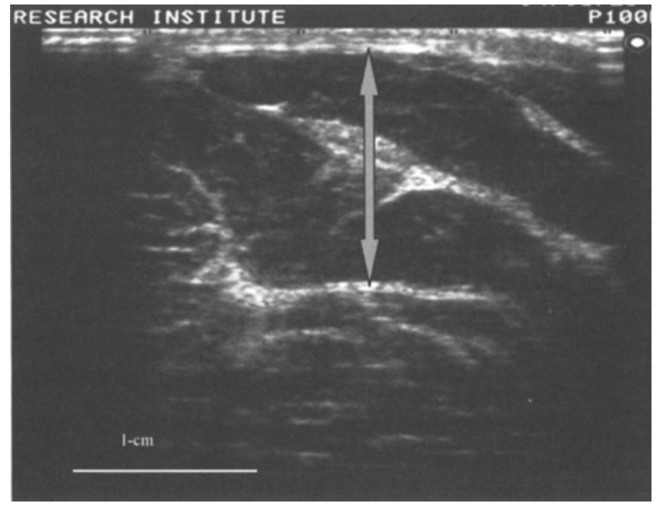

Fig 1: Ultrasound image of the extensor digitorum communis (EDC). Ultrasound-measured muscle thickness of the EDC is indicated by the arrow.

(Fig 1). We have previously determined that the CV of this measurement was 1.0% (Abe *et al.* 2004). To measure SDFT thickness, 3 longitudinal and 1 transverse images were obtained from the mid-metacarpal region of the SDFT (level 2B), as described previously by Kasashima *et al.* (2002). The distance between the border of the SDFT using a longitudinal image was taken as SDFT thickness. The CV of this measurement was 2.6%.

Statistical analysis

Results are expressed as mean ± s.d. for all variables. Statistical analyses were performed by a 2-way analysis of variance (ANOVA) with repeated measures (Group [KAATSU-walk and Control-walk] x Time [before and after training]). Serum somatotropin was analysed with a 2 x 4 ANOVA with repeated measures. *Post hoc* testing was performed by a Fisher's least significant differences test. Statistical significance was set at $P<0.05$.

Results

Foreleg girth, muscle thickness, and tendon thickness

After 2 weeks of training, ultrasound-measured muscle thickness of the UL-FDP increased (3.5%; $P<0.05$) in the KAATSU-walk

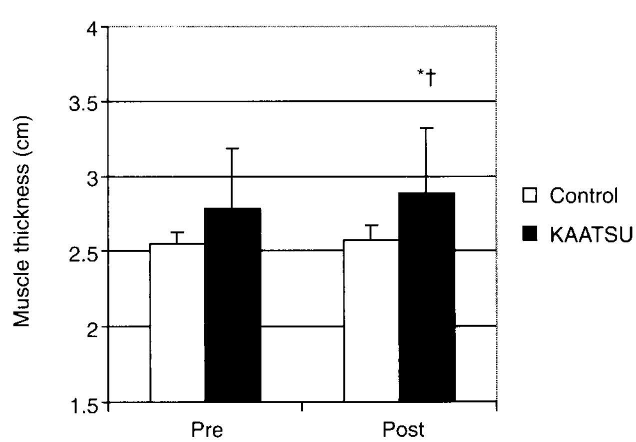

*Fig 2: Ultrasound-measured muscle thickness of the extensor digitorum communis (EDC). Measurement site for the EDC was midway between the olecranon and the accessory carpel bone, taken before and after training. *$P<0.001$ between groups. †$P<0.05$ pre- vs. post training.*

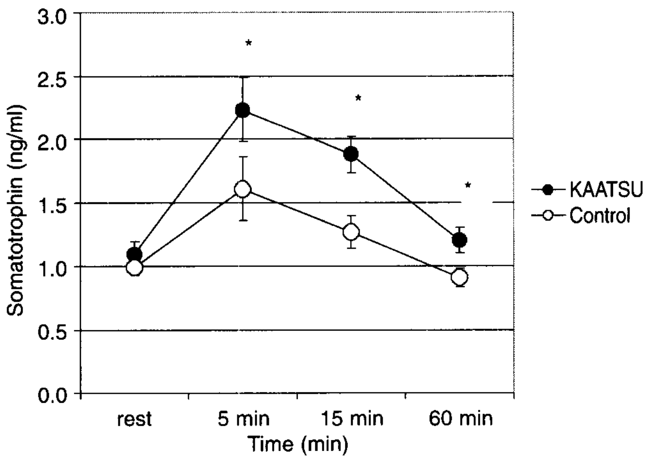

*Fig 3: Plasma somatotropin concentrations (ng/ml) at rest, 5, 15, and 60 min post exercise. All data are means ± s.e. *P<0.05 between groups.*

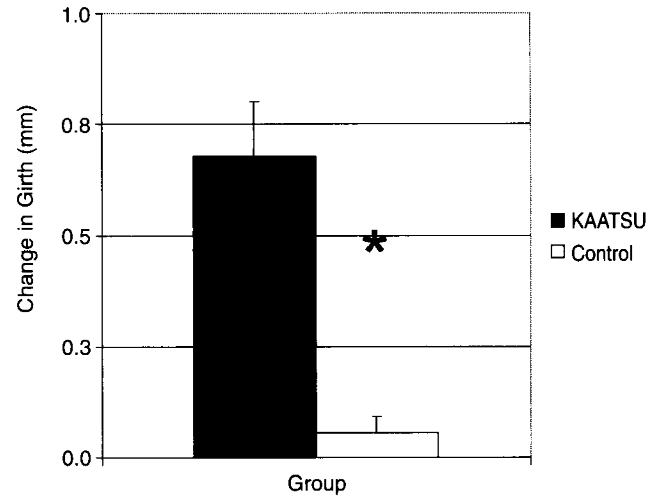

*Fig 4: Girth measurements of the horse forelimb following acute KAATSU-walk or Control-walk. Girth measurements were taken at the midpoint between the olecranon and the accessory carpal bone. *P<0.001 between groups.*

group but did not change (0.7%; P>0.05) in the Control-walk group (Fig 2). The change in muscle thickness was larger (P<0.05) in the KAATSU-walk group compared with the Control-walk group. After daily exercise, mean change in forelimb girth was higher in the KAATSU-walk group than in the Control-walk group (Fig 4). There was no change (P>0.05) in the superficial/deep digital flexor tendon thickness following 2 weeks of training in either the KAATSU-walk or Control-walk groups.

Serum somatotropin

Serum somatotropin concentration was elevated from prior to the start of exercise to 5 min (P<0.05), 15 min (P<0.01), and 60 min (P<0.05) after exercise in the KAATSU-walk group. The serum somatotropin peaked to 2.2 times that of the pre-exercise level at 5 min after acute walking in the KAATSU-walk group (Fig 3). In the Control-walk group, somatotropin rose at 5 min after acute walking, but this increase was not significantly different from pre-exercise levels. The increase in somatotropin seen in the KAATSU-walk group was larger (P<0.05) than in the Control-walk group at all measured time points.

Discussion

The primary finding of the present study was that KAATSU-walk training produced significantly increased muscle thickness of the EDC in horses. The 3.5% increase in foreleg muscle thickness was similar to results seen in man following KAATSU-walk training (Abe *et al.* 2006). In addition, KAATSU-walk training produced a rise in plasma somatotropin following an acute training session. Acute plasma somatotropin increased 120% in the KAATSU-walk group, double the increase seen in the Control-walk group. The magnitude of change in somatotropin was not as large as that seen in man (Takarada *et al.* 2000a) but was sufficient to produce muscle hypertrophy.

KAATSU-style training has consistently produced growth in skeletal muscle in man (Kawada 2005) using low-intensity resistance training (Takarada *et al.* 2000b; Abe *et al.* 2005a) or even walking (Abe *et al.* 2006). In the present study, we demonstrate for the first time that KAATSU training produces muscle hypertrophy in horses similar to that produced in man. Previously, we demonstrated that KAATSU training was safe for use in horses and did not result in any blood flow abnormalities, suggesting that inflammatory adverse effects in the lower limb (such as laminitis) would be unlikely (Abe *et al.* 2004). Therefore, KAATSU training and the resultant muscle hypertrophy should produce a beneficial therapeutic and/or rehabilitative value for equine athletes.

The exact mechanisms of KAATSU-induced muscle hypertrophy are not fully understood. However, it has been suggested that such factors as somatotropin, insulin-like growth factor-1 (IGF-1), and/or other myogenic regulatory factors (i.e. myostatin) are believed to play important roles in KAATSU-training-induced muscle hypertrophy (Abe *et al.* 2005a; Kawada 2005). Previous work has demonstrated that KAATSU training causes increases in somatotropin (Takarada *et al.* 2000a) and IGF-1 (Abe *et al.* 2005a) while down-regulating myostatin, a negative regulator of muscle hypertrophy (Kawada and Ishii 2005). In the present study, plasma somatotropin concentration doubled following acute KAATSU-walk training, and while this increase was not as robust as that seen in man (Abe *et al.* 2005b), it was sufficient to produce muscle hypertrophy. Neither IGF-1 nor myostatin was measured in the present study, so it is uncertain how these systems would be affected in horses; further work is warranted.

Owing to the short duration of our training study, however, there were no changes in SDFT thickness (Fig 5). Previous studies have shown equivocal results regarding the effects of exercise on SDFT hypertrophy. Given the fact that changes in tendon take longer than skeletal muscle, it is possible that training longer than 2 weeks may prove to be beneficial for equine tendon. KAATSU training is also believed to cause beneficial changes in bone remodelling. Human clinical studies have shown that femoral head avascular necrosis or bone atrophy was improved following KAATSU resistance training (Inoue *et al.* 2002). In addition, a recent study in man demonstrated that twice-daily KAATSU training (as opposed to the once-daily regimen used in the present study) for 3 weeks led to significant increases in the bone turnover

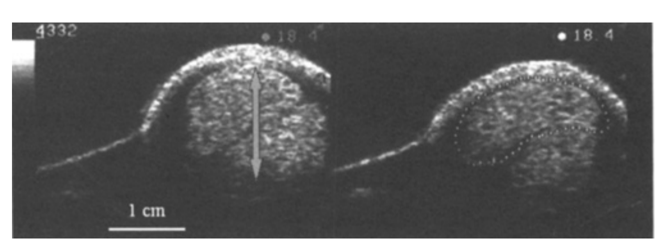

Fig 5: Ultrasound image of the superficial digital flexor tendon of the horse forelimb.

marker bone-specific alkaline phosphatase (BAP) (Beekley *et al.* 2005). BAP levels are believed to reflect osteoblastic activity and have been used as a marker of bone formation (Gundberg 2000). Whether KAATSU-walk training would alter BAP or cause any significant changes in bone remodeling in horses is unknown, but given the seriousness of orthopaedic problems in horses, this would seem an important line of research to pursue.

In conclusion, these data demonstrate that KAATSU-walk training resulted in significant increases in plasma somatotropin and a small, but statistically significant increase in muscle hypertrophy. There, it is possible that KAATSU-walk training may provide significant therapeutic/rehabilitative value in horses.

Manufacturers' addresses

[1]Kaatsu Master, Sato Sports Plaza, Tokyo, Japan.
[2]Active Equine Growth Hormone ELISA, Diagnostic Systems laboratories, Inc., Webster, Texas, USA.

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