

## Acute Compartment Syndrome Model Using Turkey Tibialis Cranialis Muscle

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**INTRODUCTION:** Acute Compartment Syndrome (ACS) is a common and severe condition following extremity trauma, which every orthopedic surgeon has experienced at one time or another. In animal models of ACS, the most common method to induce ACS is to infuse a solution such as saline or an albumin suspension directly into the muscle compartment. Although small-animal models such as rats and rabbits have some advantages such as cost effectiveness and the availability of biological analytical tools and reagents, they are technically challenging in terms of ACS induction, clinical examination including ultrasound, and functional evaluations. Therefore, large animals such as dog and pig are more frequently employed. Recently, we developed a novel turkey model for musculoskeletal research. For modeling ACS, turkeys have several advantages to compared to other species. (1) Turkeys have large leg muscles that have a similar anatomy and structure as human legs for they are 2-pedal walking animal and are amenable to clinical ultrasound and functional evaluations. (2) Turkeys have thin skin with little fat in the subcutaneous tissue and compartment. We would like to report the usefulness of this model for ACS study.

**METHODS:** We used a total of 32 Bourbon Red Heritage turkeys (equal ratio of males and females; Little Bend Heritage Farm, LLC, Chatfield, MN), about 1 year old (10-15 kg) and skeletally mature. Turkeys were divided into 4 groups (n=8, Fig.1): Groups 1 and 2 had the ICP maintained at 50 mmHg for 2 and 6 hours, respectively. During the 50mmHg maintenance period, ultrasound shear wave elastography (SWE) was measured using an ultrasound system (SuperSonic® MACH™ 30, Hologic, Marlborough, MA) every half hour at the same time points as when ICP measurements are taken. Groups 3 and 4 had a fasciotomy performed after 2 and 6 hours, respectively, of ICP at 50 mmHg. Once each animal was under general anesthesia, the left or right leg, selected randomly, was prepared for ACS creation. The tibialis cranialis (TC) muscles, which corresponds to tibialis anterior muscle in human, were identified with ultrasound by extending the ankle and feet. A slit catheter and intra-compartmental pressure monitor (Stryker, Kalamazoo, MI) were inserted into the center of the muscle to measure and monitor the inter compartment pressure (ICP) continuously throughout the procedure. A 19-gauge needle with a catheter was inserted into the deep anterior compartment and 5% solution of chicken albumin was be infused continuously with a rotating IV pump (Harvard pump, Harvard Apparatus, South Natick, MA). Infusion speed was adjusted based on the desired inter compartment pressure (ICP). SWE was simultaneously analyzed at the following six testing points: 0 (before infusion), 10, 20, 30, 40, and 50 mmHg. Each pressure level was maintained for ten minutes, and three SWE data points were collected during each stabilized pressure interval (Fig.1). During ICP elevation and maintenance period, the SWE was measured at the same time hourly to correlate the ICP. During follow-up, SWE was used to measure muscle elasticity daily during the first week after the procedure, every two days during the second week, and then weekly thereafter until sacrifice. Then, the relation between ICP and SWE was investigated. The half of the turkeys was euthanized 6 weeks after ACS. Just before euthanizing, isometric tetanic muscle force (ITF) test to the TC muscle was performed under general anesthesia. The tibial nerve was fully exposed bilaterally, and the nerve branch of the common peroneal nerve was identified. A miniature bipolar electrode (Harvard Apparatus, Holliston, MA) was attached to the peroneal nerve to stimulate TC muscles. The distal TC tendon was released and attached to the force transducer (MDB-0.5, Transducer Techniques, Temecula, CA). The contraction force of TC muscle was measured under stimulating the peroneal nerve at 10V, duration 0.4s and delay 2ms with preload at 100N. Additionally, we collected pedobarographic and kinetic data of turkeys weekly using portable walkway system (emed-x1, Novel, Munich, Germany). Parameters including contact area (cm<sup>2</sup>) and contact force (N) were calculated using designated software (Novel Projects, Novel, Munich, Germany).

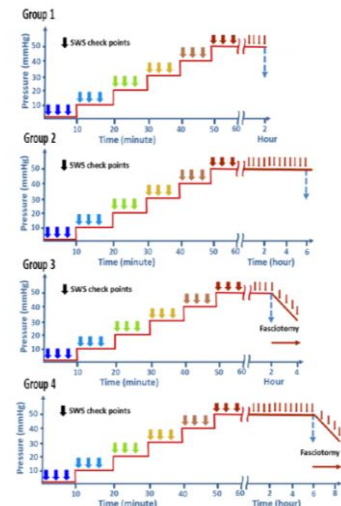


Fig 1. Experimental setup, showing four groups of turkeys and the planned pressure, SWE checkpoints, and fasciotomy (in groups 3 and 4).

**RESULTS:** In all group, values of the SWE (kPa) were significantly correlated with the ICP (mmHg) during ACS ( $R^2=0.91$ ). In group 3, average values of SWE (kPa) of ACS side were significantly higher (29.81kPa at 1<sup>st</sup> week, 32.17 at 2<sup>nd</sup>, 27.04 at 3<sup>rd</sup>, 26.79 at 4<sup>th</sup>) than the control one (20.69kPa at 1<sup>st</sup> week, 18.06 at 2<sup>nd</sup>, 17.60 at 3<sup>rd</sup>, 20.11 at 4<sup>th</sup>) during 1-4 weeks after making ACS. Comparing between with fasciotomy and without fasciotomy among group 2 and 4, average values of SWE (kPa) with fasciotomy (20.41kPa) were significantly lower than those without fasciotomy (31.41kPa) 6 weeks after ACS. Comparing between 2hrs and 6hrs of the infusion term in group 1 and 2, average values of SWE (kPa) of 6hrs (24.34 kPa) were significantly higher than those of 2hrs (18.11kPa) during 1-2 weeks after making ACS. ITF test results showed that average construction forces of TC muscle of ACS side with fasciotomy (1562N in 2hrs ACS, 2761 in 6hrs ACS) were not significantly changed to those of control side (1028N in 2hrs ACS, 1732 in 6hrs ACS) in group 3 and 4. However, average forces of ACS side without fasciotomy (1768N in 2hrs ACS, 2029 in 6hrs ACS) were significantly lower than those of control side (2714N in 2hrs ACS, 2399 in 6hrs ACS) in group 1 and 2. Comparing between with and without fasciotomy among turkeys after 6hrs infusion for ACS, average forces with fasciotomy (2761N) were significantly higher than those without fasciotomy (2029N). Walkway analysis showed that both average contact area and force of 2nd and 3rd digit was increased in the last 50% of each step 1 and 2 weeks after ACS. Comparing between 2hrs and 6hrs of the infusion term, average time-integral contact forces of the ACS side of 6hrs (24.3N) were significantly higher than those of 2hrs (18.8N).

**DISCUSSION:** The ultrasound SWE measurements showed significant and high correlations with the actual ICPs. The SWE measurements were capable of sensitively detecting changes in the order of 10 mmHg with ICP, and may well be used as an alternative to the invasive ICP measurement currently used in clinical practice as a diagnostic method for ACS. The results of the ITF tests and walking way analysis showed that the infusion made both the construction power and the function of TC muscle be reasonably reduced in line with the variation of situations that can occur in real clinical practice, such as differences in the term ACS occurred and the subsequent fasciotomy. In terms of developing an experimental model using turkeys, our model may be useful as an inexpensive bipedal large ACS model.

**SIGNIFICANCE/CLINICAL RELEVANCE:** We have confirmed that turkeys are potentially useful for a novel, bipedal and inexpensive large animal model for ACS using ultrasound measuring, ITF test and walkway analysis. Additionally, the ultrasound SWE measurement is potentially alternative method for diagnosing ACS to invasive ICP measurement penetrating needle to the muscle.