Experiences with sheep as an animal model for shoulder surgery: Strengths and shortcomings

A. Simon Turner, BVSc, MS, Dipl ACVS, Ft. Collins, CO

Sheep (and goats) are a convenient large-animal model for rotator cuff repair because of availability, ease of handling and housing, animal cost, and acceptance to society as a research animal. Tenotomy of the infraspinatus tendon and subsequent reattachment to the proximal humerus is useful to address the biomechanical, histologic, and biochemical processes of rotator cuff repair. Detaching this tendon and immediately reattaching it does not represent the clinical picture but serves as a relatively rapid way to screen different suture anchors, suture patterns, scaffolds, growth factors, and other biologics or a combination of these treatments to enhance the healing process. To minimize spontaneous reattachment and reproduce a chronic rotator cuff injury, the end of the tendon can be covered and then reattached 4 weeks later if bone-to-tendon healing is to be evaluated. This chronic model is useful to understand the biology (degree of muscle atrophy and fatty infiltration) of rotator cuff muscles as well as innovative methods of repair. Close-stall confinement is required during the convalescence in acute and chronic studies. Ultrasound in the awake animal can be used to monitor gap formation and tissue organization. Sheep have also been used to determine whether capsular healing after plication is equivalent to open capsular shift. (J Shoulder Elbow Surg 2007;16:158S-163S.)

Skeletally mature female sheep are rapidly becoming a convenient large-animal model for a variety of orthopedic problems, including rotator cuff repair. This is because of availability, ease of handling and housing, animal cost, and acceptance to society as a research animal. Although the anatomy of the shoulder of quadrupeds is quite different than humans, tenotomy of the infraspinatus tendon of goats and sheep, and subsequent reattachment to the proximal humerus, is useful to address the biomechanical, histologic, and biochemical processes of rotator cuff repair. Sheep have been selected because the similarity of the infraspinatus tendon to the human supraspinatus tendon. Although the sheep infraspinatus tendon is not intraarticular, there is a bursa under the tendon, and therefore, the repair has some contact with synovial fluid that lubricates the bursa. The bursa likely does not have the volume of synovial fluid that is contained in the shoulder joint. Therefore, the repaired tendon has some characteristics of a healing tendon in a milieu of synovial fluid. In most cases, retrieval surgery harvests robust tendon-like scar material that forms between the retracted tendon stump and the bone (gap scar formation). This is not analogous to the human condition in which gaping or failure to heal occurs in rotator cuff repair.

This tendon also useful for in vitro studies because it is relatively easy to prepare for biomechanical testing owing to its size. Surgery on one shoulder is well tolerated in sheep. The Institutional Animal Care and Use Committee (IACUC) may only allow the surgery to be performed on one forelimb, but some IACUC’s may allow both. Preemptive and postoperative analgesia is essential to all animal studies.

SURGICAL APPROACH

Shoulder surgery in sheep is performed under general anesthesia with the animal in lateral recumbency. An open approach to the shoulder is used because arthroscopic repair would be impractical. A gently curving skin incision is made over the point of the shoulder and then deepened through the subcutaneous tissues. The skin is reflected and the fascia of the brachialis muscle is incised. The acromial head of the deltoideus muscle is incised. The acromial head of the deltoideus muscle will become visible. A plane of dissection is established along the cranial edge of the deltoideus muscle and the muscle reflected caudad. Important anatomic structures of the shoulder joint in sheep are displayed in Figure 1. The insertion of the infraspinatus tendon is identified, and a curved forceps placed under the tendon. The tendon is incised along its attachment to the greater tuberos-
Figure 1 A, Intraoperative view of the surgical approach to the right tendinous insertion of the infraspinatus muscle in the sheep. The acromial head of the deltoideus muscle is retracted. B, Anatomic dissection of a right shoulder shows the location of M. teres minor. C, Further dissection of a right shoulder shows the synovial bursa and the footprint of M. infraspinatus. D, Deeper dissection of a right shoulder shows the humeral head and joint capsule. E, Bony anatomic landmarks of the right sheep shoulder.
ity of the humerus (Figure 1, A and B). The footprint of the tendon is approximately 10 mm × 20 mm (Figure 1, C). Immediately under the tendon is a synovial bursa that does not communicate with the shoulder joint (Figure 1, C). The shoulder joint is not entered in this approach (Figure 1, D).

At this point, a Hall air drill and burr are used to create a bed of bleeding bone, and if a bone trough is to be used, it is prepared at this stage. Orthopedists new to this model will be surprised and pleased at how dense sheep bone is compared with adult or elderly human bone. Predrilling of the bone is necessary for suture anchor insertion. Acute and chronic studies of infraspinatus reattachment will be discussed.

ACUTE STUDIES

Clearly, detaching the ovine (or caprine) infraspinatus tendon and immediately reattaching it does not represent the clinical picture. It does, however, serve as a relatively rapid way to screen different suture anchors, suture patterns, scaffolds, growth factors, and other biologics, or a combination of these treatments, to enhance the healing process (Figure 2). Survival times frequently used are 6 and 12 weeks. Group sizes are typically 12 animals if mechanical testing and histology are end points. Within each group, 9 animals are allocated to mechanical testing and 3 for histology.

Some of the earlier studies using sheep (and goats) have evaluated different methods of tendon reattachment. St. Pierre et al. used goats to evaluate tendon healing to cortical bone compared with a cancellous trough. They evaluated the repairs histologically and biomechanically and found the tendon-to-bone healing process quite similar.

Other acute studies using sheep have looked at different suture anchors (Figure 3), a mixture of bone morphogenetic proteins, suture patterns, low-intense pulsed ultrasound (Figure 4), and swine small intestinal submucosa.

CHRONIC STUDIES

The repair of a chronic, massive, rotator cuff injury is a challenge to the shoulder surgeon. This has prompted the search for a clinically relevant animal model to evaluate methods to enhance repair and understand the biology behind the chronic tear. Initially, delayed repair of the released infraspinatus tendon was not recommended because of the difficulty in distinguishing scar tissue from normal tendon
at the time of reattachment. As a rule, animals are known to heal rapidly, with abundant neovascularization and fibrous tissue ingrowth; sheep are no exception.

In later studies, methods to cover the end of the infraspinatus and minimize spontaneous reattachment were refined. To reproduce a chronic rotator cuff injury, Coleman et al1 covered the end of the infraspinatus with Gore-Tex (PRECLUDE; W. L. Gore & Associates, Flagstaff, AZ; Figure 5). The sheep were reoperated on 6 weeks later. The PRECLUDE-covered tendon was located, isolated, and reattached to the lateral tuberosity of the humerus. The tendon had retracted an average of 2.6 cm (range, 1.9-2.9). In that study, some sheep were reoperated on at 18 weeks. These animals had irreparable tendons because of excessive retraction, which were secured using a synthetic scaffold as a bridge.

To aid in identification of the detached infraspinatus tendon after long periods of detachment, Gerber et al4 performed an osteotomy of the greater tuberosity of the humerus. To prevent spontaneous healing, the end of the tendon was covered with a 5-cm-long silicone rubber tube. They also found fatty infiltration and muscle atrophy in proportion to the amount of musculotendinous retraction. Therefore, the chronic detachment model in sheep is also useful to understand the biology (degree of muscle atrophy and fatty infiltration) of rotator cuff muscles as well as innovative methods of repair. The chronic model gives the surgeon a better understanding of the timing of the repair and the temporal aspects of healing.1,4,11

Figure 5 Identification of the infraspinatus tendon covered with Gore-Tex (PRECLUDE, W. L. Gore & Associates, Flagstaff, AZ) in a chronic model.1

These long-standing chronic models (detached for periods greater than 8 weeks) are more useful to evaluate bioimplants (eg, collagen scaffolds) rather than attempted reattachment to the bone. In sheep, we currently recommend infraspinatus detachment and covering, and then reattachment, as soon as 4 weeks if bone-to-tendon healing is to be evaluated. If the ability of a bioimplant, scaffold, autologous platelet-rich fibrin matrix, a growth factor or a combination of these to bridge a large gap is to be evaluated, the reattachment surgery can be scheduled 8 weeks after the detachment and covering surgery.

Some of the earlier problems using the sheep model of rotator cuff repair were related to suture rupture caused by failure to protect the repair from full weight bearing. Because slinging was poorly tolerated in sheep and not permitted by our IACUC, a rubber ball was placed under the hoof of the involved limb to see if this restricted limb movement, and this was removed at 5 weeks.8

Lewis et al8 looked at the effect of immobilization on infraspinatus healing using a modified Mason-Allen suture. They found that there was no difference between the treatment groups for load-to-failure and stiffness, so this cannot be recommended as a method of immobilization. Rather, we have reverted to close-stall confinement during the convalescence.

In summary, it is virtually impossible to control the postoperative loads on the repaired tendon, and there are no data to suggest that the ball on the hoof actually leads to diminished weight bearing. Furthermore, other researchers with experience with this model suggest that the sheep fires the muscles as it resists and “fights” the ball on the hoof. Therefore, this is not a method to “immobilize” the repaired shoulder. Because sheep and goats are large, heavy animals, they likely fire the shoulder girdle muscles immediately on standing up after surgery.

The high loads imposed by weight-bearing result in some separation of the repair in this model. The gap between the distal end of the transected tendon and the proximal humerus can be identified on the ultrasound images in the awake but restrained sheep, without costly general anesthesia, to monitor progression or lack of healing (Figure 6).

Although many of these repairs fail to heal by tendon-to-bone healing, the model is still useful to study the effect of chronic tendon detachment on muscle atrophy and fatty infiltration. The model can also be used to study the effect of various devices and factors on decreasing the rate of healing of primary repairs for them to remain intact or the biologic effects on the formation and maturation of tendon-like scar formation in a preclinical model using a rotator cuff surgical site.

Other issues with this model that should be mentioned are that the forelimb is weight bearing and there is no clavicle, a less-developed acromion, and no coracoacromial arch. These differences between
the sheep (goat) and humans are probably not as important as the issue mentioned above.

An important factor related in this, and likely other large-animal models, is that all of the repairs detach to some degree. The intervening gap fills in with fibrous scar tissue, unlike humans where a gap typically forms, which may be due to the extraarticular nature of the repair. As a result, this is a model of tissue formation in a gap. As such, the model may have real advantages as a “tissue engineering” approach to tissue formation, but likely it does not evaluate direct healing between tendon and bone.

One distinct disadvantage of the model is a lack of species-specific probes and reagents for sheep (eg, antibodies), which limits the ability to conduct detailed histologic analyses such as immunohistochemistry and in-situ hybridization.

**Glenohumeral Instability Assessment**

The shoulder of the sheep can be used to evaluate the response of the glenohumeral joint capsule. Obrzut et al. used explanted sheep shoulders to evaluate the effect of radiofrequency energy on the length and temperature properties of the capsule. Sheep have been used to determine whether capsular healing after plication is equivalent to open capsular shift (Figure 7). As determined from histologic appearance, there was no difference in the 2 techniques.

**SUMMARY**

Research facilities around the world are continuing to gain more experience with sheep as a practical and economic large-animal model for various therapies of the shoulder joint, particularly the enhancement of bone-to-tendon healing. In addition, researchers are continuing to refine the model and learn more about its limitations. Studies of growth factors at different doses and stem-cell therapy, in combination with different scaffold materials and their configurations, are likely to dominate this exciting field of orthopedic research.

**REFERENCES**


