

Clinical Application of an Acellular Biologic Scaffold for Surgical Repair of a Large, Traumatic Quadriceps Femoris Muscle Defect

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abstract

Many battlefield injuries involve penetrating soft tissue trauma often accompanied by skeletal muscle defects, known as volumetric muscle loss. This article presents the first known case of a surgical technique involving an innovative tissue engineering approach for the repair of a large volumetric muscle loss.

A 19-year-old Marine presented with large volumetric muscle loss of the right thigh as a result of an explosion. The patient reported muscle weakness with right knee extension, secondary to volumetric muscle loss, primarily involving the vastus medialis muscle. This persisted 3 years postinjury, despite extensive physical therapy. With all existing management options exhausted, restoration of a portion of the lost vastus medialis muscle was attempted by surgical implantation of a multi-layered scaffold composed of extracellular matrix derived from porcine intestinal submucosa. The patient had no complications, was discharged home on postoperative day 5, and resumed physical therapy after 4 weeks. Four months postoperatively, the patient demonstrated marked gains in isokinetic performance. Computer tomography indicated new tissue at the implant site.

This approach offers a treatment option to a heretofore untreatable injury and will allow us to improve future surgical treatments for volumetric muscle loss.

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Loss of muscle mass due to combat injury is a significant problem facing the military. Penetrating soft tissue injuries involving volumetric muscle loss are common on the battlefield and often result in severe cosmetic deformities, chronic muscle weakness, and debilitating loss of function. Management of large areas of volumetric muscle loss can be challenging because patients desire both performance improvement and cosmetic enhancement. Successful replacement of functional muscle tissue following large volumetric muscle loss is one of the more difficult tasks for the reconstructive surgeon due to the lack of available treatments.

Current clinical management practices include autologous tissue transfer, vascularized or free muscle flaps, and externally applied orthotic equipment to augment reserve muscle function. Muscle flaps are primarily used to provide soft tissue coverage, and generally do not restore strength. Free muscle transplantation has resulted in successful reconstruction of larger defects in the forearm¹ and elbow.² However, these procedures are highly specialized, and limited success has been reported in only a small, select patient population. These procedures are not applicable to large volumetric muscle loss. Autologous tissue transfers, either pedicle or free flap, are limited by donor site morbidity, a limited supply of tissue, and the immediate perioperative concern for infection, flap failure, and failure to restore contractile function.

Regenerative medicine and tissue engineering therapies offer a possible solution. Tissue engineering has been increasingly used as an alternative in other specialties for tissue replacement. Omori et al³ developed a tissue scaffold from Marlex mesh for repair of the larynx and trachea in 4 patients. Biologic scaffolds have been associated with recruitment of endogenous progenitor cells,⁴⁻⁶ rapid degradation and replacement with host tissue,^{7,8} and the presence of muscle tissue in cardiac,⁹

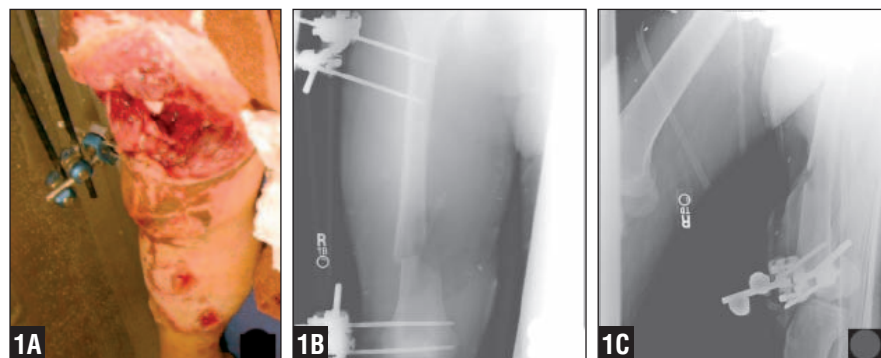
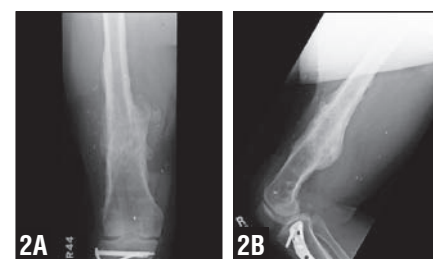


Figure 1: Image of initial presenting injury (A). An external fixator is placed for femur fracture, which eventually heals but the patient continues to have muscle weakness of his right quadriceps due to volumetric muscle loss. AP (B) radiograph on arrival at Landstuhl Regional Medical Center, Germany. **Figure 2:** AP (A) and lateral (B) radiographs after bony healing and hardware removal.



esophageal,¹⁰ and lower urinary tract application.¹¹⁻¹³

This article presents a case of the first application of a biologic scaffold composed of porcine small intestinal submucosa extracellular matrix for the purpose of regenerating functional muscle tissue in the extremity following a large volumetric muscle loss sustained from military trauma.

CASE REPORT

A 19-year-old Marine presented with multiple injuries from an explosion. Ten inpatient admissions totaled 231 days. A majority of time involved treatment of his right femur fracture, which was successfully repaired via internal fixation, autologous bone graft, and latissimus muscle flap. He sustained an open fracture to his right lower extremity (Figure 1). Although this open fracture eventually healed (Figure 2), the associated large volumetric muscle loss of the surrounding quadriceps muscle, primarily the vastus medialis, was the long-term clinical challenge. After stabilization of the bony defect, treatments for quadriceps defect included placement of a Latissimus dorsi free flap and strength training to maximize residual muscle function.

For the first 1.75 years following injury, physical therapy was problematic and inconsistent due to the injury severity and multiple

complications. The next 1.75 years included consistent, rigorous physical therapy designed specifically to address the deficit in quadriceps muscle strength.

Three and a half years postinjury, the patient reported muscle weakness with right knee extension secondary to the large volumetric muscle loss. A secondary report was dissatisfaction with the lack of a normal contour to the thigh. Functional impacts included difficulty descending stairs and low endurance during ambulation. The patient consented to participate in the Innovative Surgical Treatment procedure at our institution. The Innovative Surgical Treatment is part of normal surgical practice that uses FDA-approved products, benefits the patient, respects patient autonomy to provide informed consent, and does not involve a comparison of treatments.¹⁴ Because of the novelty of the specific application, the process established by the United States Army Institute of Surgical Research adds peer review of the surgical plan and preplanned assessment of outcomes to ensure process improvement.

The Innovative Surgical Treatment involved the surgical implantation of a custom-manufactured implant composed of porcine small intestinal submucosa extracellular matrix biologic scaffold material (Restore, DePuy Orthopaedics).

edics, Inc, Warsaw, Indiana) within the muscle defect area. At this time, his Biodex testing peak torque production at 90°/second extension measured 28% of his contralateral side.

The small intestinal submucosa-extracellular matrix device consisted of 10 layers of small intestinal submucosa vacuum pressed into a strong multilaminar sheet (5×7×0.02 cm). The small intestinal submucosa material has been well characterized and consists of structural proteins including collagen I, III, IV, V, and VI; fibronectin; glycosaminoglycans; and functional molecules such as vascular endothelial growth factor, basic fibroblast growth factor, and transforming growth factor beta among others.¹⁵⁻¹⁹ The material is not chemically crosslinked, is vacuum dried, and is terminally sterilized by electron beam radiation.

The implant was hydrated by soaking in a bowl of sterile water at room temperature 10 minutes prior to use. The right thigh was prepared and a 15-cm curvilinear incision was made lateral to the skin scar on the thigh. This incision was carried down to the vastus lateralis, exposing the posterior fascia. The muscle belly was dissected free and lifted anteriorly from the fascia via blunt and sharp dissection. The dissection was carried medially allowing adequate visualization and mobilization of the muscle belly. A 12×10 cm pocket was created and 5 sheets were placed flat into this space. A 10-French (French Catheter Scale) three-quarter-fluted Blake Drain was then placed above the implant, and the skin incision was closed in 2 layers, followed by a sterile dressing. The procedure was well-tolerated with no complication. The drain was removed 24 hours postoperatively, and the patient was discharged home on postoperative day 5. Four weeks postoperatively, he resumed physical therapy.

The patient began regular physical therapy 1.75 years prior to this surgery. The physical therapy sessions were conducted 3 times per week and involved stationary cycling (15 minutes/session), weight training involving squats with free weights, and leg presses at 3 sets of each exercise, and the resistance was adjusted to a level that could be repeated 10 times (10 repetitions max). Additionally, leg extensions were routinely performed on the Biodex Sys-

tems 3 isokinetic dynamometer (Biodex Medical Systems Inc, Shirley, New York) consisting of isokinetic quadriceps strengthening at low and high speeds. In addition to physical therapy, the patient engaged in regular cycling. At 4-week follow-up, the patient returned to physical therapy. He was ambulatory with no assistance.

The routine use of the Biodex System 3 prior to testing ensured that the patient was thoroughly familiar with the testing procedure. All Biodex testing was conducted under the supervision of a physical therapist. The same isokinetic testing protocol was used each testing session. Tests were conducted 1 week preoperatively and 16 weeks postoperatively. Testing involved 5 repetitions/trial at 90° per second. Each time the patient was tested, the

same range of motion, test velocities, repetitions, and seat positions were used.

The volumetric muscle loss resulted in a dramatic loss of isokinetic muscle function. Preoperatively, the values for peak torque, total work, and average power for the involved limb were: 41 ft-lbs, 89 ft-lbs, and 38 watts, respectively. At the same time, the values for the uninvolved limb for peak torque, total work, and average power were: 147 ft-lbs, 492 ft-lbs, and 177 watts, respectively. Postoperatively, the patient demonstrated marked improvement in all measured parameters (Figure 3). In contrast, values for the uninvolved contralateral limb displayed declines in many of the measured parameters postoperatively.

Pre- and post-computed tomography (CT) comparison demonstrated the presence of soft

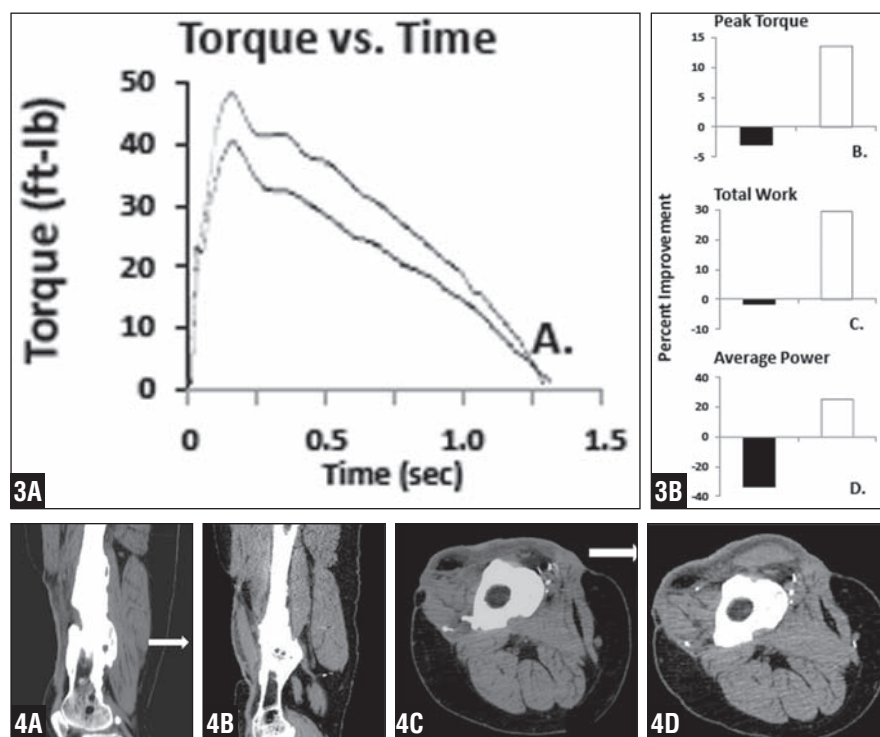


Figure 3: The torque curves for leg extension pre- and postoperatively as illustrated by the letter A (A). The curves represent the averaged data for 5 repetitions. These data were used to determine the variables presented in B to C. The increase in peak torque and average power (torque×time) can be seen in this figure. The torque curves for the uninvolved limb are not shown. All values B-D are expressed as the percent improvement $[(\text{postoperative value} - \text{preoperative value}) / \text{post surgical value}] \times 100$. Note that at the same time the treated leg improved, the uninvolved limb underwent a reduction in the measured variables (B). **Figure 4:** CT image of injured right lower extremity. Axial CT scan through the right lower extremity at 2-mm slice thickness with sagittal and coronal reformatted images. A and C are sagittal and coronal images (respectively) obtained 5 months prior to implantation of the extracellular matrix. B and D are sagittal and coronal images (respectively) obtained 9 months after implantation. Arrow indicates the presence of 1.9×4.9×9.4 cm-tissue not present pre-implantation.

tissue measuring 1.9×4.9×9.4 cm (Figure 4). Magnetic resonance imaging (MRI) comparison was not feasible because of retained foreign body fragments.

At the time of final testing, the patient reported that he felt his cycling and walking endurance had improved and he was able to walk up and down stairs more easily and with greater stability.

DISCUSSION

Extremity injuries represent 63% of primary diagnoses for admission²⁰ and are the primary source of disability in 69% of patients medically retired after combat injury.²¹ To what extent large volumetric muscle loss contributes to this number is unknown, but complex, large skeletal muscle extremity defects after military trauma are not uncommon. The current management options for volumetric muscle loss of the magnitude displayed in this patient are limited. Muscle flaps of sufficient magnitude to return function to the affected musculature are not feasible due to the size of the donor tissue required and limited ability to create functional constructs. These facts dictate a need for the development of new treatment options. To this end we have reported on the first case involving a tissue engineering approach for the treatment of large volumetric muscle loss.

The 36-week post-implantation CT scan revealed new tissue with dimensions 1.9×4.9×9.4 cm corresponding to product sheets that were implanted. Badylak et al¹⁶ demonstrated that after 12 weeks the originally implanted small intestinal submucosa could not be identified using a monoclonal antibody for porcine small intestinal submucosa in a dog skeletal muscle defect model. After 36 weeks, the continued presence of soft tissue is permanent and unlikely to be postoperative edema or retained xenogenic extracellular matrix.

Clinically, this patient currently has a palpable soft tissue mass that addresses the secondary complaint of abnormal con-

tour. In other parts of the body (eg face and forearm), restoration of more normal appearance may be a primary goal and this extracellular matrix approach warrants consideration. Although MRI is the ideal diagnostic modality for soft tissue imaging, patients that suffer volumetric muscle loss in the military often have retained metal fragments (Figure 4), a contraindication to MRI. However, the presence of new tissue was significant enough to be detectable with current CT protocols. Confirmation of the composition of the soft tissue would require histological analysis of biopsy samples.

The improvements in isokinetic performance are encouraging and were consistent with the results of the CT scan. The validity of assessing quadriceps muscle dynamics using the Biodex isokinetic dynamometer is well accepted. The trial-to-trial and day-to-day reliability of the Biodex system has been previously evaluated.²² The Biodex protocol used was identical every time. Additionally, the patient had almost 2 years of routine use of the Biodex for testing and physical therapy; thus the post surgical improvement cannot be attributed to a learning effect. Because contralateral measurements decreased postoperatively (likely a detraining effect) the data as gain over presurgical value are presented to ensure a conservative analysis. Thus we are confident that the results indicate a true gain in strength postoperatively. However, the data reflect the combined contribution of all of the quadriceps muscles; it is impossible to determine the extent to which the uninvolved synergists contributed to the improvements.

Subjectively, the patient perceived gains in strength and endurance and has requested additional extracellular matrix implantation. This subjective data may be a placebo effect but it is consistent with the isokinetic results. Of particular interest is the gain in power and work measurements. The speed chosen, 90°/second, is similar to that of routine daily living such as rising from a chair and negotiating stairs. The

finding that these values were improved is concordant with the patient's subjective self-assessment.

Small intestinal submucosa-extracellular matrix has been used in >1 million patients with both positive and, in select patients, mixed results.²³ The Restore Orthobiologic Implant is an extracellular matrix product that is composed of matrix derived from the porcine small intestinal submucosa. Its composition and ultrastructure promote angiogenesis, host cellular infiltration, and site specific remodeling with restoration of normal structure. While the radiological and functional data, as well as the patient subjective assessment, are encouraging, the fact is that muscle function remains well below that of the uninvolved leg. The inability to completely restore function underscores the need for continued animal and human research in muscle engineering. Further efforts are required to more fully restore function and further optimize tissue engineered solutions.

A review of muscle engineering is far beyond this article^{24,25}; however, in the most general terms muscle engineering can be divided into cell-based and scaffold-based approaches. Cell-based approaches rely on the seeding of a single cell type,²⁶ or differentiated stem, or progenitor cells, onto a supporting scaffold material.²⁷ Within this approach are many variations in terms of types of scaffolds and cell sources, however most involve the use of a bioreactor to control the biochemical, nutritional, thermal, and mechanical environment of the developing construct.²⁷ This approach has been successful in developing small muscle constructs capable of producing force.^{27,28} However, the constructs have been small; and limited by adequate vascularization and innervations, and are not currently available for clinical use.

The second, alternative approach is a scaffold-based approach (used in the current case), which exploits the ability of the biological scaffold to recruit endogenous

progenitor cells to the site of injury,⁴⁻⁶ rather than depending on the addition of exogenous cells. In addition to the ability to attract progenitor cells, biological scaffolds undergo rapid degradation and replacement with host tissue^{7,8} and have been associated with the presence of muscle tissue in repair applications for skeletal muscle²⁹; cardiac³⁰; esophageal³¹; and lower urinary tract.¹¹ Another distinction of this approach is that it relies on the recipient's body as the bioreactor, depending on it to supply the appropriate biochemical, nutritional, thermal, and mechanical environment.

Finally, a third approach involving the introduction of exogenous stem cells into a previously implanted scaffold has recently shown promise for supporting the growth of new muscle cells.^{32,33} 

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