EnFix™ Implants for Enthesis Repair

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Unmet Clinical Need

There are 500,000 rotator cuff surgeries performed annually in the USA[1]. Operative treatment is still far from perfect, with repair failure rates of 20% to 94% reported in the literature. Ratcliffe et al's[2] review of the literature reported failure rates for rotator cuff repair as assessed by MRI or ultrasound imaging. Their results are shown in Table 1 below.

Tear Size	Failure Rate ± SD	Range
Small to Medium (1 – 3 cm)	22 ± 7%	10 – 40%
Large (3 – 5 cm)	46 ± 21%	10 – 95%
Massive (2 or more tendons)	58 ± 12%	37 – 76%

Table 1. Rotator Cuff Failure Rates from Ratcliffe et al[2].

Objective evaluation of the repairs has demonstrated a high rate of incomplete healing and gap formation at the interface between the tendon and the bone, i.e., at the enthesis. Failure of adequate tendon-to-bone ingrowth places the repair at risk for re-tear.

In the uninjured state, the tendon-bone interface, or enthesis, has a fibrocartilage transitional region that exhibits gradations in cell phenotype, matrix composition, tissue organization, and mechanical properties. These natural gradations facilitate the effective transfer of load between two materials of greatly differing stiffness by reducing the potentially damaging stress concentrations that would otherwise arise at their interface. Numerous studies[3–6] have shown that the enthesis has a poor healing potential and a weaker scar tissue generally forms. The resultant repair has a lower strength[3,7]. Enhanced biologic repair at the interface between tendon and bone would be a major advance.

Over several decades, biologic tendon scaffolds derived from human and animal tissues have been generated, and synthetic scaffolds have been manufactured from absorbable and non-absorbable polymers to reinforce and replace tendons and ligaments. These patch products are placed on the tendon and do not seek to affect healing of the enthesis.

The importance of enthesis repair was highlighted in a 2017 NIH / NIAMS hosted roundtable on Innovative Treatments for Enthesis Repair[8]. Preclinical research has investigated several strategies to generate an enhanced biologic repair including growth factors, demineralized bone matrix, tissue engineering, cell therapy, hyperbaric oxygen therapy, shockwave, and low intensity pulsed ultrasound. While some of these studies have shown promising results, the translational research requirements to commercialization are often significant.

Demineralized Bone Matrix and Enthesis Repair

The seminal work of Marshall Urist's initial discovery of bone morphogenic proteins (BMP)s in 1965[9] came from his observation that bone implants created from demineralized bone matrix (DBM) had the ability to induce the formation of new bone in experimental animals. He hypothesized that DBM contained a factor that could stimulate bone growth, and subsequent experiments led him to isolate and characterize the first BMPs. Urist's discovery has had a significant impact in orthopedics and has led to the development of new treatments for bone and joint injuries and diseases. BMPs are used clinically in spinal fusion surgery and other orthopedic procedures to promote bone healing and regeneration. Allograft DBM has also been shown to be a potent biomaterial with significant clinical usage, as a bone void filler and graft material in spine surgery[10].

There have been some studies of the effect of demineralized bone matrix products on enthesis repair reviewed by Hexter et al[11] in 2017. Sundar et al[12] produced strips of demineralized allogeneic bone to use in a sheep model of tendon enthesis healing. Strips of DBM 15 x 30 x 2-3mm were interposed between tendon and bone and held in place using suture anchors. They saw early failures in the control group that were not seen in the treated group. Histological analysis at 12 weeks demonstrated reformation of the enthesis in the DBM treated group but not in the control group. Smith et al[13] studied rotator cuff healing in a dog model using a demineralized cancellous sponge loaded with PRP. They demonstrated improved histology, MRI scores, and repair strength at 12 weeks. Lovric et al[14] showed that DBM powder introduced into the bone tunnel of an ACL repair in a rodent model demonstrated increased graft strength at 4 and 6 weeks. Heuberer et al[15] injected DBM powder into the tendon footprint of sheep and showed less scar tissue and a more physiologic enthesis morphology at 4 weeks.

Interpretation of the results of the DBM studies is hampered by the variation in DBM materials used, ranging from xenogenic to allogenic, from demineralized cancellous sponge to demineralized cortical bone and demineralized bone powder.

The Solution

Tetrous' EnFix implants are the first-to-market, procedure-specific implants manufactured using patented Demineralized Bone Fiber (DBF) technology focused on enthesis healing to address stubbornly high failure rates in rotator cuff repair surgery. Tetrous was spun out of TheraCell, a regenerative medicine company in 2019 to further its patented DBF technology for application in sports medicine.

The EnFix products are allogenic tissue products that conform to the Food and Drug Administration's (FDA) regulations governing human and cellular tissue-based products (HCT/P) according to 21 CFR Part 1271 and Section 361 of the PHS Act. The processes used to manufacture EnFix products were designed to cause minimal changes to the allograft tissue and to maintain the osteoinductive potential. The products are 100% cortical bone and contain no additives or excipients.

While DBM is a potent biomaterial, in its most-used form as a powder its handling characteristics are lacking and moreover it lacks osteoconductivity. Excipients used to make DBM powder easier to handle are deficient in that they can contain up to 70% extraneous binding materials that have no beneficial value as biomaterials or to overcome the lack of osteoconductivity. Demineralized bone fiber technologies offered a means to improve osteoconductivity as superior putties but had no means to yield shaped allograft. Tetrous utilizes a second-generation fiber technology that overcomes many of the limitations of earlier manufacturing methodologies and provides a means of producing procedure specific shaped products while simultaneously yielding highly osteoinductive and osteoconductive properties. The patented technology also facilitates retention of the nanotopography of the collagen fibrils. DBF implants produced using this technology have been used in an estimated 150,000 spine procedures since 2016.

Two versions of the EnFix implant have been produced, EnFix RC™ and EnFix TAC™. The EnFix RC implant is designed to be used in conjunction with suture anchors. It is suture anchor agnostic and works well with most widely used threaded suture anchors of sizes 4.5mm - 6.5mm.

The EnFix RC design rationale required use of DBF to provide optimal biologic performance while also making it easy for the surgeon to use with minimal disruption to the surgical technique. This was achieved by molding and manufacturing the DBF fibers into a "Top Hat" shape wherein the shaft of the implant sits in the awl hole used for suture anchor insertion to allow cells and other endogenous local factors to wick up from the subchondral bone to the top surface of the implant that sits at the interface between bone and tendon. The top of the implant is 8.5mm in diameter, allowing optimal spacing of suture anchors, and the peg portion is 13mm long.

Figure 2. The EnFix RC implant

Use of EnFix RC does not require the surgeon to change their surgical technique and adds only 2 minutes to the surgery time. The EnFix RC implant is placed into the awl hole created for the suture anchor and then held in place using the suture anchor. The implant is tapered with four ribs designed to resist rotation during suture anchor insertion. An additional benefit of the usage of the EnFix RC implant is that the peg portion of the implant enhances fixation of the suture anchor into bone, in the same manner as TheraCell's award winning Fiber Anchor enhances pedicle screw fixation in compromised pedicles.

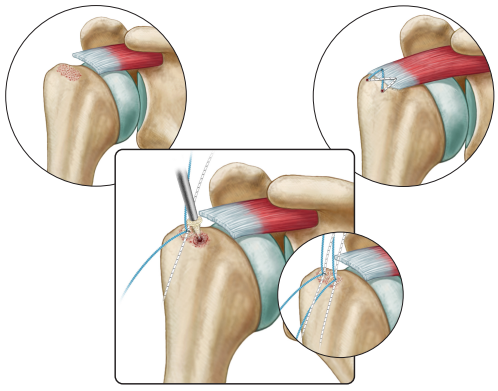

Figure 3. EnFix RC surgical placement

The EnFix TAC product is available in two forms and is designed to be used remote from a suture anchor making it suitable for use in conjunction with all-suture suture anchors or where the surgeon wishes to promote enthesis repair remote from the site of suture anchor insertion. The EnFix TAC is available in two designs; TAC-O has an 8.5mm diameter top, while the TAC-T product has a 4mm x 10mm top. The implant is designed to be placed between the medial and lateral rows. The implant dimensions are shown in Figure 4.

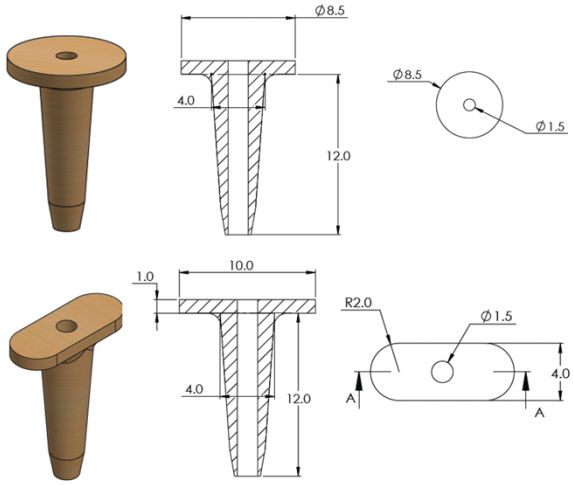

Figure 4. Dimensions of the EnFix TAC products

Preclinical Studies

The EnFix technology has been evaluated in multiple preclinical studies. Traditional demineralization techniques apply acid treatment after the fiber is formed, etching the surface nanotopography (surface features) away. Tetrous' unique patented and proprietary process demineralizes bone struts first and then cuts/cleaves fibers along the bone's long axis, thus maintaining their important nanotopography, and creating long and strong fibers[16]. See Figure 5 below.

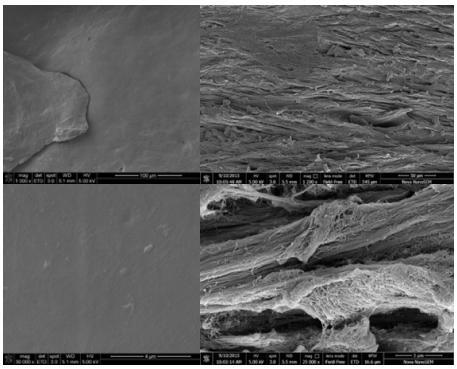

Figure 5. Conventional DBM / DBF surface topography, left panels, Tetrous DBF right panels. Conventional DBM processing results in a smooth surface due to powdered bone being demineralized in acid while Tetrous DBF retains natural nanotopography as a result of demineralization prior to fiber cutting.

One design feature of the implant is the ability to allow cells and bone marrow to be wicked up and proliferate through the implant to the enthesis. Mercury Intrusion Porosimetry was used to characterize the implant and showed a porosity of 39% with an average pore size of 160 µm. In Figure 6 below, comparison is shown to two enthesis repair products that use electrospun fabrics. These materials have a very small pore size that is less than 10 microns and too small for cellular transport.

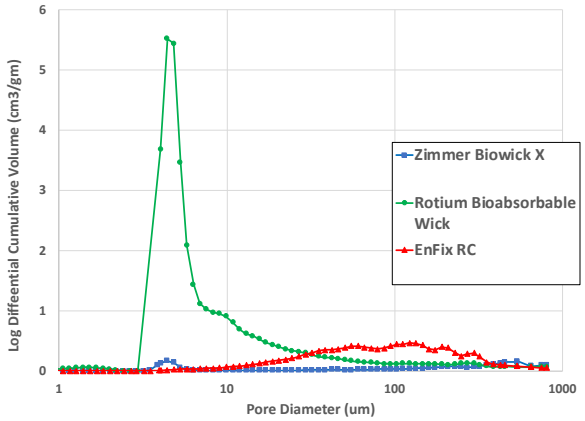

Figure 6. Pore diameter distribution for EnFix compared to competitor products.

Production of implants with intricate geometries required the development of a proprietary patented water assisted injection molding (WAIM) process that allows fibers to be suspended as a slurry and injected into shaped molds. A further process, designated Formlok™, causes the shape to be retained even in a wet environment such as is experienced in arthroscopy to control implant integrity. Figure 7 below shows the effect of water immersion on the integrity of the implant.

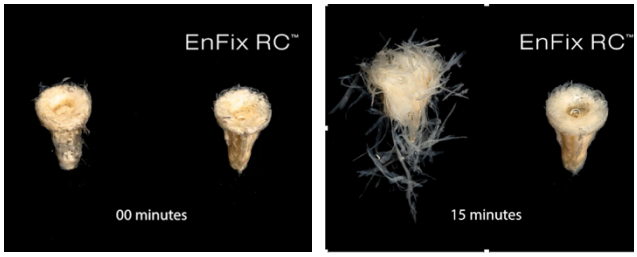

Figure 7. The implant on the left is untreated and loses shape rapidly on immersion in water. Implant to the right is Formlok™ treated.

As the only additive to the process is water, the manufacture of the DBF and the formed implants conform with the requirements of minimal tissue manipulation as defined and regulated by the FDA.

Evaluation of suture anchor fixation in the EnFix RC implant was performed in a laboratory model. Sawbones 10pcf cellular rigid polyurethane foam (Pacific Research Laboratories, WA, USA) is a well-accepted analog for osteopenic cancellous bone and is specified for screw testing in ASTM standards and FDA Guidance documents[17]. A Mark 10 motorized test stand with a 1500N Force gauge was used for pull out testing with a pull out rate of 20mm/min. A Zimmer Biomet Quattro 5.5mm PEEK suture anchor was either placed directly into the Sawbones foam block or into an EnFix RC implant that was placed into the Sawbones foam block. The maximum pull out force data are shown in the figure below, with standard deviation, n=5. The EnFix RC is shown to provide a modest improvement in fixation.

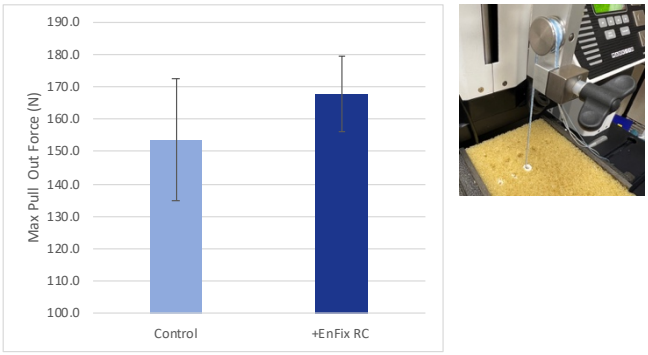

Figure 8. Photograph shows test set up for suture anchor pull out. Chart shows maximum pull out force observed.

DBF has become one of the most commonly used bone grafting materials and the ability of the Tetrous DBF to promote bone healing was evaluated in a critical sized distal femoral defect in a skeletally mature rabbit model. The results demonstrated rapid bone formation, growing from the outside of the defect towards the center at two weeks with complete remodeling of the DBF into bone seen at four weeks. Bone formation is through a process of endochondral ossification[9].

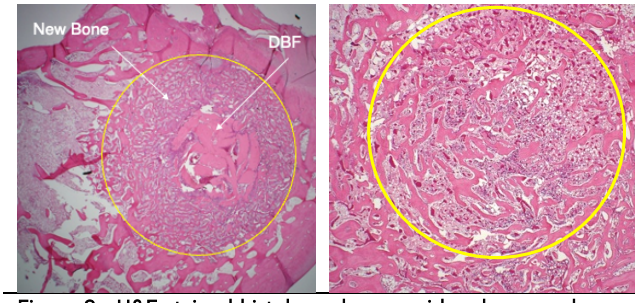

Figure 9. H&E stained histology shows rapid early woven bone formation at 2 weeks, left and 4 weeks, right.

In a sheep model of enthesis repair presented as Poster # 105 at the 2023 AOSSM meeting, a DBF sheet was placed at the interface between tendon and bone. Histology showed enthesis reformation at 12 weeks with Sharpey's Fibers in the treated group, but not the control.

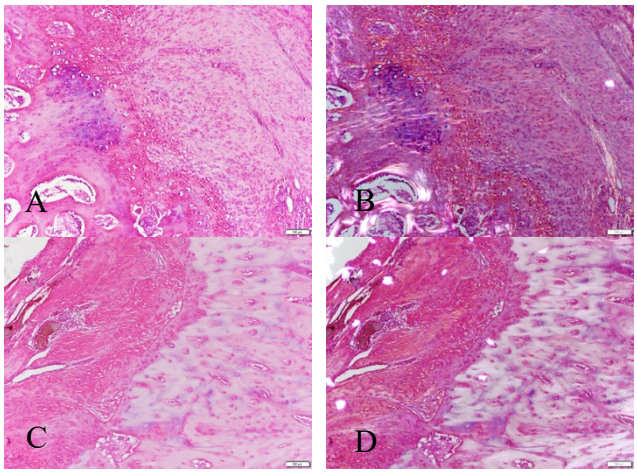

Figure 10. H&E histology at 12 weeks under normal light for the DBF treated (A) and control (C) revealed an active interface with some residual DBF and a reforming enthesis. Polarized light confirmed Sharpey's fibers in the DBF treated group not present in the controls (B vs D).

In a separate study, 6.5mm diameter bone screws were placed into fiber sleeves mimicking the peg portion of the EnFix RC implant and placed into skeletally mature sheep distal femoral condyles. The study showed that new bone formation occurred around the screw facilitated by the DBF fibers.

These data are shown in Figure 11 below. New bone formation can be seen at four weeks, with some residual DBF, while at 12 weeks all of the DBF has remodeled into new woven bone in the areas between the screw threads.

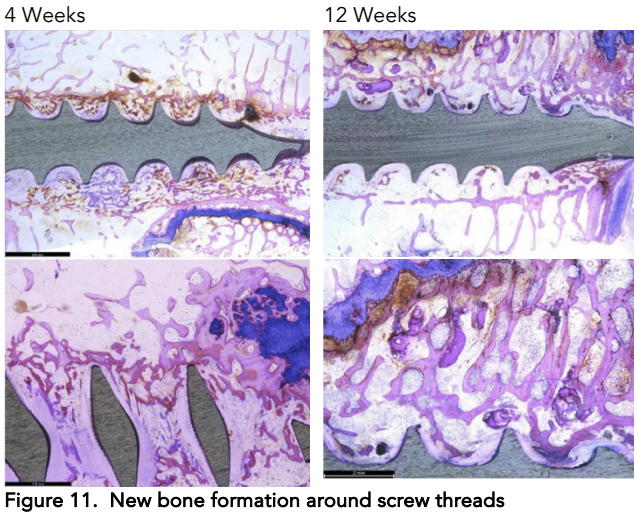

Figure 11. New bone formation around screw threads

A rabbit model of tendon repair was investigated using the infraspinatus tendon. Two bone tunnels were placed using a

1mm K-wire lateral and medial to the infraspinatus tendon insertion footprint. The infraspinatus tendon was sharply dissected from its insertion. Modified Kessler 3-0 sutures were placed through the tendon and then reattached by passing the two free ends of the suture through the bone tunnels using a straight mayo needle. In the treatment group a DBF sheet approximately 1mm thick and 5mm wide was placed between the tendon and bone. In the control group the tendon was reattached without the DBF sheet.

Tensile testing of the repaired rotator cuffs was performed and DBF and non DBF repairs compared at 6 and 12 week timepoints. No. 1 Ethibond sutures were passed through the infraspinatus tendon, and interfaced to an MTS 858 Bionix Testing Machine (MTS, Eden Prairie, MN, USA). The humerus was secured in a jig. The infraspinatus tendon–humerus complex was positioned to allow tensile loading in the longitudinal direction of the infraspinatus tendon. The repair sutures were cut prior to mechanical testing in order to isolate the testing of the healing interface alone. Specimens were preconditioned for 5 cycles of loading and unloading with 5% strain of the initial length at a cross-head speed of 6 mm/min, and then loaded to failure at a speed of 6 mm/min. The results of this testing are shown in Figure 12 below. The cuff repairs deploying the DBF implant required greater force (21% at 6 weeks and 26% at 12 weeks) to detach the tendon from the bone than the control.

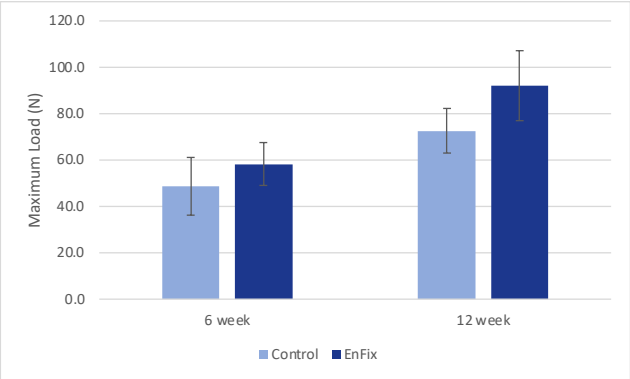

Figure 12. Maximum load of the infraspinatus tendon repair.

Pre- and Post- Surgical MRI Clinical Assessment

MRI assessment of repairs using the EnFix RC and EnFix TAC is being conducted on patients at 3 months and 6 months post op. A 6 Month post-op MRI is provided as an example in Figure 13. The MRI demonstrates excellent healing of the supraspinatus tendon repair at the greater tuberosity footprint with clear tendon-to-bone ingrowth. The MRI demonstrates high quality coverage of the greater tuberosity footprint by the repaired tendon similar in morphology to what a native tendon would look like. The only evidence that a tear has been repaired, other than the bone anchors themselves, is the mild intermediate T2 signal in the tendon substance. Note that there is little to no T2 signal between the tendon and the adjacent footplate bone, which suggests a robust tendon-bone enthesis. The EnFix RC shows successful integration into the surrounding bone, with nearly no visible evidence of the EnFix RC such as marrow edema, cystic change or adverse localized soft tissue reaction. The suture anchor is well seated in the bone and is easily visible with no artifact obscuring the tendon insertion.

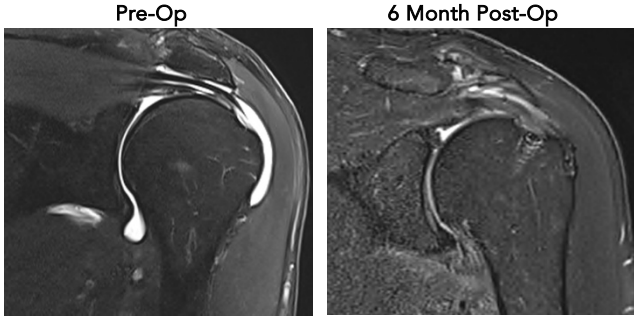

Figure 13. Example of MRI

Patents / Intellectual Property

The demineralized bone fiber technology used in Tetrous products is licensed exclusively for use in sports medicine from TheraCell, Inc., an Isto Biologics Company. In addition to this and independently, Tetrous has its own patents covering the EnFix products and methods of manufacture and use. The granted US patents applicable to the products are provided in Table 2 below. Other patent applications, in the USA and other jurisdictions, have been filed for additional coverage for products to treat rotator cuff and other entheses.

Products	US Patents
EnFix RC EnFix TAC-O EnFix TAC-T	US 9,486,557 US 9,572,912 US 11,660,373 US 11,759,548

Table 2. US Patents

EnFix™, EnFix RC™, EnFix TAC™, Tetrous™ and "It's all about the Enthesis"™ are trademarks of Tetrous, Inc.

Bone Textile™ and FormLok™ are trademarks of TheraCell, Inc.

Conclusions

The need for a means of improving the outcomes of rotator cuff repair are clearly demonstrated by the clinical data. These results have not been impacted by the improvements in suture anchor design, suture or surgical technique. Demineralized bone matrix materials are a potent orthobiologic material with significant usage in spinal

surgery. The literature on demineralized bone matrix materials for enthesis repair suggests that they may have some efficacy for this application. Tetrous has shown that it is possible to fabricate demineralized bone fiber into a consistent product that can be easily introduced into the enthesis without impacting current surgical technique. Preclinical and initial clinical data suggest that the DBF material is capable of reformation of the enthesis generating a repair that is stronger than the control scar tissue. The DBF in the EnFix products makes new bone through a process of endochondral ossification. It is hypothesized that this, being the same process that results in enthesis formation in embryo, is what allows the new bone to reform the enthesis in healing to the tendon. Clinical and preclinical studies have shown that tendon cannot re-attach to bone other than with scar tissue. EnFix allows bone to reattach to tendon with a physiologic enthesis.

Acknowledgements

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