Functional Repair of Rabbit Gastrocnemius Tendons Using Carbon Fibers

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The ability of carbon fibers to form a secure anastomosis with soft tissue will have an important bearing on any future clinical uses. A secure anastomosis in rabbit Achilles tendons has been observed for up to 12 weeks after operation using a lockingweave suture, without immobilization. Impaired function occurred from slippage of the carbon fibers, which began by six weeks after operation, and which increased progressively at 13 and 42 weeks after operation. Satisfactory results were obtained using a modified Bunnell suture (with immobilization) in which the free ends of the carbon fibers were glued together with bone cement. In another study, carbon fibers that were weaved through intact rabbit Achilles tendons did not attach to the tendons during the postoperative time interval studied (18 weeks). It is possible, although difficult, to obtain a functional repair of the rabbit Achilles tendon using carbon fibers.

The use of carbon fibers in ligament repair has attracted interest because of their reported ability to induce oriented collagen produc tion.³ The fibers have low shear strength, and this obviates knot tying because the fibers will break when the knot is cinched. This creates problems of anchorage in both soft tissue and bone, but it may be a key factor in their role in ligament repair. The carbon fibers initially serve as a scaffold for the growth of new collagen. If, thereafter, they gradually break, the new tissue will be subjected to load, allowing it to hypertrophy. Animal studies have been performed,` and extensive clinical experience in humans has been acquired outside the United States, but more information regarding the nature of the tissue response to carbon fibers is needed. Before performing such studies, it was necessary to develop a reliable method of soft-tissue fixation that would result in a functional long-term repair of a severed tendon. The purpose of this study is to determine the best method of attachment to soft tissue, and to examine the process of repair of carbon fiber sutured tendons.

METHODS

White, female, New Zealand rabbits (3-5 kg) were used in all studies. After induction of anesthesia (intravenous pentobarbital, 30 mg/kg), the gastrocnemius/plantaris tendon complex of one leg was exposed through a longitudinal incision and the tendon sheath was opened in line with the incision. The plantaris muscle was transsected proximally at the musculotendinous junction and distally at the calcaneus and excised, exposing the two gastrocnemius tendons. A 1 cm segment of each tendon was removed.

Group 1: In 16 rabbits, the defect was repaired with carbon fibers, using a locking-weave anastomosis both proximally and distally.' Both gastroc-

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Received: May 3, 1985.



FIG. 1. Bunnell-weave repair (Group 2) of rabbit gastrocnemius tendons.

nemius tendons were incorporated in the repair as a single tendon. The limb was not immobilized.

Group 2: In 20 rabbits, the carbon fibers were passed through a transverse drill hole in the calcaneus, then crossed through the distal end of the tendons immediately proximal to the calcaneus and passed longitudinally through the 1-cm gap into the proximal ends of the tendons. A Bunnell-type repair

was used proximally (Fig. 1), and the fibers exited from the tendons immediately distal to the musculotendinous junction. The ends of the carbon fibers were twisted together and then glued with bone cement (Howmedica). Both gastrocnemius tendons were incorporated in the repair as a single tendon. A long-leg cast (20° flexion at the knee joint and 10° plantar flexion at the ankle joint) was applied for three weeks.

Group 3: Eleven rabbits underwent a unilateral sham procedure in which the tendons were resected but no implants were used. In six of these animals, a long-leg cast (20° flexion at the knee joint and 10° plantar flexion at the ankle joint) was applied for three weeks. Five animals were not immobilized.

Group 4: In 20 rabbits, carbon fibers were passed through the intact tendons bilaterally. One end of a carbon-fiber bundle was placed in the distal por tion of the tendon by passing the carbon fibers from one side of the tendon to the other, and then back again. The other end of the bundle was passed through the sheath, and placed under the skin.

In all four groups, the tendon sheath and skin were closed with 4-0 vicryl sutures using a running stitch. Sacrifice times and numbers of animals sacrificed at each time period for the various experimental groups are given in Table 1.

CARBON-FIBER IMPLANTS

The implant (Plastafil, Johannesburg, South Africa) consisted of 10,000 loosely twisted carbon-fiber strands, each approximately 7 m in diameter. To

	Ultimate Tensile Strength (newtons)										
	Group 1			Group 2				Group 3			
Postoperative Time (weeks)	n	Operated Side	Control Side	n	Operated Side	Control Side	n	Operated Side	Control Side		
0	4	51.2 ± 13.8	182.4 ± 33.4	6	140.1 ± 20.5	305.1 ± 31.1					
3							1	137.9	273.6		
4				2	154.8 ± 8.0	312.3 ± 2.2	2	149.0 ± 50.3	313.6 ± 37.8		
6	4	238.4 ± 14.7	293.6 ± 23.6	3	180.2 ± 35.1	314.9 ± 7.1	4	222.8 ± 22.7	302.5 ± 10.7		
8				3	187.7 ± 33.4	292.7 ± 25.8	2	164.6 ± 50.3	314.9 ± 23.6		
10				3	224.6 ± 63.6	326.0 ± 24.5					
13	4	$197.5 \pm 42.2*$	301.6 ± 22.7	3	201.1 ± 49.4	321.2 ± 68.0					
15							2	143.2 ± 14.2	303.4 ± 32.0		
42	4	$182.4\pm48.5*$	378.5 ± 75.6								

 TABLE 1. Average Ultimate Tensile Strength and Standard Deviation of the Gastrocnemius System at Various Times after Operation

* Slippage and elongation of the tendons occurred in all cases

At each time period in each group, the operated side was weaker than the control side (p < 0.05).

maintain flexibility and prevent separation or unravelling of the carbon fibers during surgery, the implant was coated by dipping it in a solution of gelatin dissolved in glycerin. The implant theoretically was capable of carrying a load of greater than 1000 N, as calculated from actual tensile strength measurements on individual carbon fibers. In addition, carbon fibers coated with polylactic acid were used in one study involving some of the rabbits in Group 4. Details regarding the polylactic acid coating are given elsewhere.²

All mechanical testing was done within one hour after sacrifice. A tensile load was applied to the muscle-tendon unit on an Instron Testing Machine (Model 1321) at a rate of 0.17 cm/sec. In Groups 1, 2, and 3, the femur and calcaneus were mounted with a hexagonal array of set screws.² Force-extension curves were recorded during each mechanical test and used to determine the ultimate tensile strength of each specimen. In Group 4, the tensile load was applied between the calcaneus and the free (proximal) end of the carbon-fiber implant. All statistical comparisons were based on the paired *t*-test.

Immediately after sacrifice, specimens containing carbon fibers were fixed in formalin, embedded, and sectioned in either paraffin (6-10 μ m) or epoxy (2-3 μ m) We could not obtain representative histologic sections because of the brittleness of the carbon fi bers and a tendency of the microtome blade to dislodge the carbon fibers from their tissue bed. Therefore, scanning electron microscopy (SEM) was used to visualize the relationship of the carbon fibers to the neotendon. Formalin-fixed specimens were dehydrated in ethanol, fractured with a razorblade in liquid nitrogen, critical point dried, mounted on aluminum studs, coated with a thin layer of gold, and examined in an SEM (AMR 1200) at an accelerating voltage of 25 kV.

RESULTS

GROUP 1

The initial strength of the locking-weave repair immediately after the procedure was 51.2 N (28% as strong as the normal tendon, p < .05) and failure occurred at the implant site (Table 1). Six weeks after surgery, the re pair had lengthened due to slippage at the attachments and the carbon fibers were covered with a thick fibrous sheath. The average strength of the operated specimens was 238.4 N (81% as strong as the specimens from the control side, p < .05) (Table 1). Failure consistently occurred through the belly of the gastrocnemius muscle in both groups of specimens.

At 13 weeks, the fibers had slipped further (Fig. 2) and the entire carbon-fiber bundle, including the portion used to make the locking weave, was embedded in a concentric cylinder of new fibrous tissue. The operated specimens were 65% (p < .05) as strong as those from the control side. By 42 weeks, there was a further decrease in muscle mass and thinning of the fibrous tissue that covered the carbon fibers (compared with 13 weeks). At both time periods, both groups of specimens failed through the muscle.

GROUP 2

The tendon maintained its normal length in all animals (Fig. 3). During the 13 weeks of the study, the repaired side never achieved more than 69% of the strength of the normal side (Table 1).

An SEM view of an eight-week specimen is shown in Figure 4. The implant was embedded in a concentric cylinder of connective tissue. A few, apparently broken, carbon fibers were located around the bundles; these were completely surrounded by masses of connective tissue. The two fiber bundles were still largely intact, with areas of apparently loose fibers, indicative of a lack of total tissue infiltration. The tendon sheath blended in with the newly formed tissue. Because all tissue inside the tendon sheath (Fig. 4B) had been removed during surgery, everything found in this area was new tissue.

GROUP 3

Following the sham procedure, a fibrotic reaction resembling that seen following carbon-fiber implants was observed, and six weeks after surgery it was at least 74% as strong as the contralateral control (Table 1). No comparison could be made with the carbon-fiber repairs because the specimens failed through muscle. There was no difference between animals that had been immobilized



Fig. 2. Carbon-fiber repair in Group 1, 13 weeks after operation. The femur gastrocnemius muscles, tendons, and calcaneus from each leg are shown. The control side is on the right.

compared with those that had not been immobilized.

GROUP 4

At sacrifice, the free end of the carbon fiber was covered with a transparent tissue sheath. The force required to pull the carbon fibers out of intact tendon did not increase with time (Table 2).

DISCUSSION

A secure anastomosis has been reported for up to 12 weeks after surgery using a locking weave suture without immobilization. 2 We found slippage of the carbon fibers had begun by six weeks after operation and increased at 13 and 42 weeks. Progressive muscle wasting resulted, and subsequent mechanical testing led to consistent failure through the muscle belly. The size of the fiber bundle (6000 fibers, 2 compared with 10,000 fibers) may have contributed to the differing results.

Satisfactory results were obtained using a modified Bunnell suture if the free ends of the carbon fibers were glued together with bone cement and the limb immobilized for three weeks. With this method, no significant slippage of the repair occurred (Fig. 3), and normal use of the foot was observed after removal of the plaster cast. Even so, the strength of the new tendinous tissue could not be determined because all specimens failed through muscle. Atrophy of the gastrocnemius muscle on the



FIG. 3. Carbon-fiber repair in Group 2 13 weeks after operation. The femur, gastrocnemius muscles, tendons, and calcaneus from each leg are shown. The control side is on the right.

experimental side was apparent in all animals, probably as a result of the operation and the immobilization.

When the tendon was not repaired, scar tissue bridged the defect by six weeks. The strength of the bridging scar appeared to be respectable, but could not be compared with that obtained with carbon fibers, because fail ure occurred through muscle and there was no way to estimate how much stronger the tendon would have been. However, the animals were unable to use the limbs due to the elongated tendon.

In one study attachment of carbon fibers to soft tissue was not impressive; the pull-out strengths of carbon fibers from intact tendons showed little change over an 18-week period (Table 2). The carbon-fiber coating, whether gelatin or polylactic acid, did not affect the mechanical strength of the attachment site (Table 2). Because the carbon fibers were passed through intact tendon, the only injury present resulted from the trauma of the procedure. Therefore, this study should be distinguished from the studies (Table 1) in which carbon fibers were used to repair a defect in the Achilles tendon. In the latter studies, the magnitude of the injury, and thus the healing potential, was significantly greater.

In conclusion, these studies show that it is possible, although difficult, to obtain a secure anastomosis between carbon fibers and the



Fig. 4A AND 4B. (A) An SEM micrograph of a cross section of normal rabbit plantaris/gastrocnemius tendon (plantans tendon on the left). The tendons are enclosed in a connective tissue sheath that is opened during surgery to allow removal of the tendons and insertion of the carbon-fiber implant. The tendon sheath is sutured closed at the conclusion of the surgery. (B) An SEM micrograph of a cross section through the carbon-fiber implant eight weeks after surgery. The two bundles of carbon fibers are completely surrounded by a dense layer of connective tissue, which blends in with the tendon sheath. Some fibers are distributed throughout the tissue. The posterior tibial vessels and nerves are apparent.

TABLE 2. Average Pull-out Force and
Standard Deviation of Carbon-fiber
Used in Locking Weave Configuration in Intact
Tendons at Various Postoperative Times

	Pull-out Force (Group 4) (Newtons)								
Postop- erative Time (Weeks)	n	Gelatin- coated Carbon Fiber	No. of Tendons Tested	Polyactic Acid-coated Carbon Fiber					
0	4	9.8 ± 4.0							
1	4	8.4 ± 8.0							
2	4	9.3±5.3							
3	4	15.1±2.7							
4	4	11.6±6.7							
5			4	13.3 ± 4.9					
6	4	11.6 ± 6.7							
8	4	12.9 ± 4.0							
10	5	22.7 ± 3.1	4	17.8 ± 4.9					
15	2	14.7 ± 1.3							
18	4	16.4 ± 5.3	4	21.0 ± 12.0					

n = number of tendons tested.

Achilles tendon of a rabbit. Success requires careful attention to detail or slippage will oc cur, resulting in tendon lengthening and mus cle shortening.

REFERENCES

- I. Amis, A. A., Campbell, J. R., Kempson, S. A., and Miller, J. H.: Comparison of the structure of neotendons induced by implantation of carbon or polyester fibres. J. Bone Joint Surg. 66B:131, 1984.
- Aragona, J., Parsons, J. R., Alexander, H., and Weiss, A. B.: Soft tissue attachment of a filamentous carbonabsorbable polymer tendon and ligament replacement. Clin. Orthop. 160:268, 1981.
- Jenkins, D. H. R., Forster, I. W., McKibbin, B., and Ralis, Z. A.: Induction of tendon and ligament formation by carbon implants. J. Bone Joint Surg. 59B: 53, 1977.
- SteYn, D. G.: An experimental study on the use of a carbon fibre prosthesis for the repair of the cranial cruciate ligament in the dog. J. S. African Veterinary Assoc. 55:23, March, 1984.
- 5. Tayton, K., Phillips, G., and Ralis, Z.: Long-term effects of carbon fibre on soft tissues. J. Bone Joint Surg. 64B:112, 1982.