

Unfavorable effect of knee immobilization on Achilles tendon healing in rabbits

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Submitted 99-02-08. Accepted 99-09-16

ABSTRACT — This study was undertaken to assess the effect of knee immobilization on the treatment of Achilles tendon rupture. After their Achilles tendons were severed, rabbits were divided into 2 groups. In Group A, only the ankle joint was immobilized. In Group B, both the knee and ankle joints were immobilized. At 4 weeks after surgery, both the ultimate tensile force and stiffness of the severed tendons were significantly greater in Group A than in Group B. In Group A, dense collagen fibers were seen in the repaired tendons, and the bundles of collagen fibers were parallel to one another along the axis of the tendons. In contrast, in Group B, dilated veins and capillaries were seen in the repaired tendons, and the proliferation of connective tissue containing collagen fibers was severely reduced around these veins and capillaries and was in general irregular and uneven. These results suggest that knee immobilization retards the healing of a ruptured Achilles tendon without suture, due to congestion and tension deprivation produced by keeping the tendon static.

Immobilization of both the knee and ankle has been the standard method of treatment for Achilles tendon rupture (Stein and Luekens 1976, Lennox et al. 1980, Fruensgaard et al. 1992). Some investigators have asserted that immobilization of the knee joint is not necessary for treatment (Beskin et al. 1987, Carden et al. 1987), and recent treatment trends are towards immobilization of only the ankle (Cetti et al. 1993, Rantanen et al. 1993, Soldatis et al. 1997). However, to our knowledge, no authors have explained why immobilization of the knee joint is not necessary for the treatment of Achilles tendon rupture. To evaluate

the effect of knee immobilization on treatment for rupture of the Achilles tendon, we examined both the mechanical properties and histology of the Achilles tendon after severance in rabbits treated with ankle immobilization with or without concomitant knee immobilization.

Animals and methods

66 mature male Japanese albino rabbits weighing about 3 kg each were used. Throughout the experiments, all rabbits were allowed a standard diet and water ad libitum. They were housed with a 12–12 light-dark cycle at 22–24 °C.

During anesthesia with sodium pentobarbital (20 mg/kg body weight, i.v.), both the Achilles tendon and plantaris tendon of the rabbits were dissected free from the surrounding tissues, and the Achilles tendon with the peritendium was severed in the middle with a scalpel. The plantaris tendon was left intact as an internal splint to keep a very short distance between the two stumps of the Achilles tendon. There was no tendon gap after the severance. The severed tendon was not sutured. Black silk markers were attached to both stumps of the severed tendon to measure the distance between the stumps.

The rabbits were divided into 3 groups: those with only the ankle joint immobilized (group A, n 28), those with both the ankle and knee immobilized (group B, n 28), and a control (sham operation) group that did not undergo severance of the Achilles tendon (n 10).

In group A, the ankle was immobilized at 60° plantar flexion by percutaneously inserting a 1.8

mm Kirschner wire from the calcaneus through the talocrural articulation to the tibia. The immobilization was augmented by a short leg cast.

In group B, the knee was immobilized at 90° flexion by percutaneously inserting a 2.4 mm Kirschner wire from the anterior aspect of the tibia through the articulation to the femur. The ankle was then fixed, using the method described in group A. Immobilization was augmented by a long leg cast.

The Achilles tendon was exposed during anesthesia with sodium pentobarbital at 1, 2, 3, and 4 weeks after surgery (n 4, 8, 8, and 8 in both group A and group B). The distances between the markers at the proximal and distal stumps were measured in all rabbits.

Then, the Achilles tendon including the os calcis was harvested for mechanical tests to determine mechanical properties, using an Autograph Model AG-B (Shimadzu Corporation, Kyoto, Japan). At the distal end, a 1.2 mm Kirschner wire was inserted in the calcaneus as an anchor perpendicular to the calcaneal body, and the calcaneus was embedded in acrylic resin. The test specimen was immersed in a saline solution bath at 37 °C for 15 minutes for temperature equalization prior to testing. The proximal end of the tendon was covered with 150 grit sand paper to avoid slipping, and held with a clamp. Mechanical tests were performed at a cross-head speed of 500 mm/min to prevent avulsion fracture of the calcaneus (Noyes et al. 1974). Simultaneously, the load-deformation curve was recorded at a chart speed of 1000 mm/min. The ultimate tensile force and stiffness (slope of the load-deformation curve over the linear range) were measured in the tendons harvested at 2, 3, and 4 weeks after surgery (n 5, 5, and 5 in both group A and group B, respectively).

For histological examination, the Achilles tendons harvested at 1, 2, 3, and 4 weeks after surgery were fixed in 20% buffered neutral formalin for 3 days, dehydrated, and embedded in paraffin. Longitudinal sections of the tendon were stained with hematoxylin and eosin.

All data are expressed as means (SD) of the groups. Statistical comparison of means was made with a two-way factorial ANOVA. Groups of data were considered significantly different at $p < 0.05$.

Table 1. Distance (mm) between the stumps of the severed Achilles tendon

Observation time (weeks after surgery)	Group A			Group B		
	n	mean	SD	n	mean	SD
1	4	4.9	1.9	4	4.5	1.3
2	8	5.3	1.8	8	4.6	1.2
3	8	6.1	0.8	8	5.6	0.7
4	8	7.3	1.0	8	6.6	1.4

Results

The distance between the stumps of the severed Achilles tendon (Table 1)

The distance between the stumps of the severed Achilles tendon increased as time passed, but there was no difference in the distance between group A and group B ($p = 0.1$).

Mechanical tests of the Achilles tendon.

Load-deformation curves are shown in Figure 1. At 2 weeks after surgery, the forces in group A and group B were about one third and one eighth, respectively, that in the control group (316 (37) N, n 10). In both group A and group B, the force increased as time passed, but there was a difference in force between group A and group B ($p < 0.0001$). The force in group A was double that in group B at 4 weeks after surgery (Table 2).

The stiffness data are shown in Table 3. The stiffness at 2 weeks after surgery in groups A and B was about three tenths and one tenth, respectively, that in the control group (101 (18) N/mm, n 10). In group A and group B, the stiffness increased as time passed, but there was a difference in the stiffness between group A and group B ($p < 0.0001$). The stiffness in group A was about 1.7 times that in group B at 4 weeks after surgery.

Histological findings in the Achilles tendon.

In the control rabbits, there was no conspicuous histological change in the Achilles tendon. The tendon consisted chiefly of bundles of collagen fibers, which ran in one direction, with rows of flattened fibroblasts between them. In group A, at 1 week after surgery, most of the gap between the stumps of the severed Achilles tendon was filled with granulation tissue, blood cells, and fibrin. At

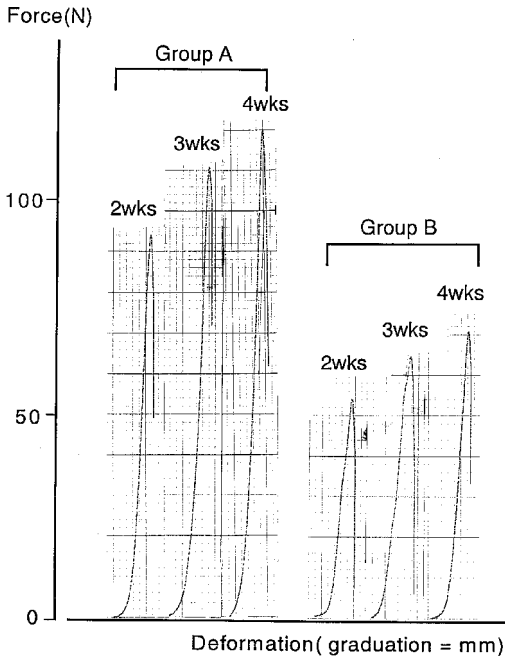


Figure 1. Typical load-deformation curves.

2 weeks after surgery, dense proliferation of collagen fibers was seen in the gap with capillaries, but the fiber bundles were not oriented along the axis of the tendon. At 4 weeks after surgery, the bundles of collagen fibers were parallel to one another along the axis of the tendon (Figure 2). However, the group A bundles were thinner the fiber bundles of the control animals.

In group B, at 1 week after surgery, most of the gap between the stumps was filled with fibrin mesh, blood cells, and a few fibroblasts. At 2 weeks after surgery, granulation tissue with dilated veins and capillaries proliferated in the gap. At 3 weeks, proliferation of connective tissue containing collagen fibers was seen with the dilated veins and capillaries. At 4 weeks, the bundles of proliferated collagen fibers ran in one direction, but the dilated veins and capillaries interrupted the pathways; as a result, tendon healing was more irregular and uneven than in group A (Figure 3).

Discussion

Appropriate tendon mobilization accelerates the healing of an injured tendon, and immobilization

Table 2. Ultimate tensile force (N) of the Achilles tendon

Observation time (weeks after surgery)	Group A			Group B		
	n	mean	SD	n	mean	SD
2	5	103	16	5	41	10
3	5	110	19	5	56	9.1
4	5	117	6.6	5	62	8.7

Table 3. Stiffness (N/mm) of the Achilles tendon

Observation time (weeks after surgery)	Group A			Group B		
	n	mean	SD	n	mean	SD
2	5	28	3.5	5	12	3.3
3	5	31	5.5	5	16	2.6
4	5	34	5.9	5	19	3.5

prevents healing (Woo et al. 1981, Gelberman et al. 1982, Murrell et al. 1994). The reasons for these effects may be as follows. Appropriate tendon movement may facilitate the diffusion of tissue fluid and place longitudinal tension on the tendon (Date 1986). Stress deprivation may change the fibroblasts from an anabolic to a catabolic state and reduce collagen mass (Amiel et al. 1990). Moreover, mechanical stress may promote the maturation of collagen through the polymerization of fibrils (Mason and Allen 1941, Becker and Diegelmann 1984) and may enhance synthesis of procollagen in the fibroblasts (Enwemeka 1991). Although the mechanism is not very clear, it seems that appropriate tendon movement and tension on the injured tendon play important roles in healing.

Our study clearly demonstrates why immobilization of both the knee and ankle retards healing of a severed Achilles tendon. Immobilization of both the knee and ankle joints disturbed the normal proliferation of connective tissue during healing, especially around dilated veins and capillaries. The dilatation of vessels means chronic congestion. Appropriate tendon movement maintains sufficient blood circulation and promotes venous

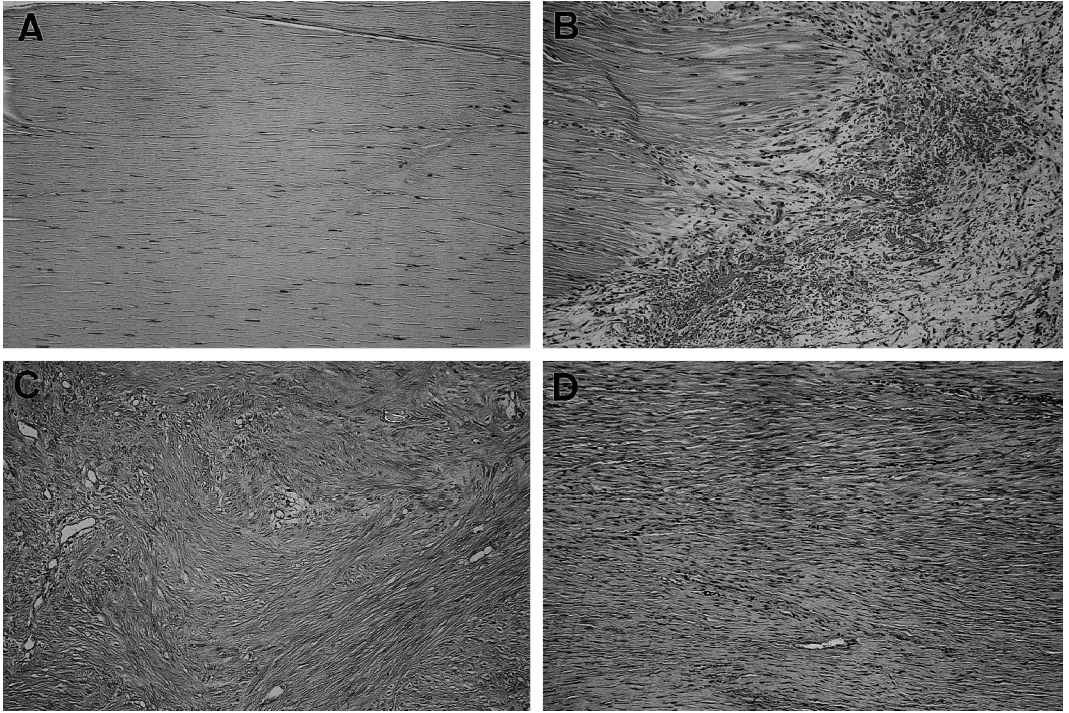


Figure 2. The Achilles tendon. H and E, $\times 100$. A. Control. B, C, and D. Specimens from group A at 1, 2, and 4 weeks, respectively, after surgery (immobilization of only the ankle joint).

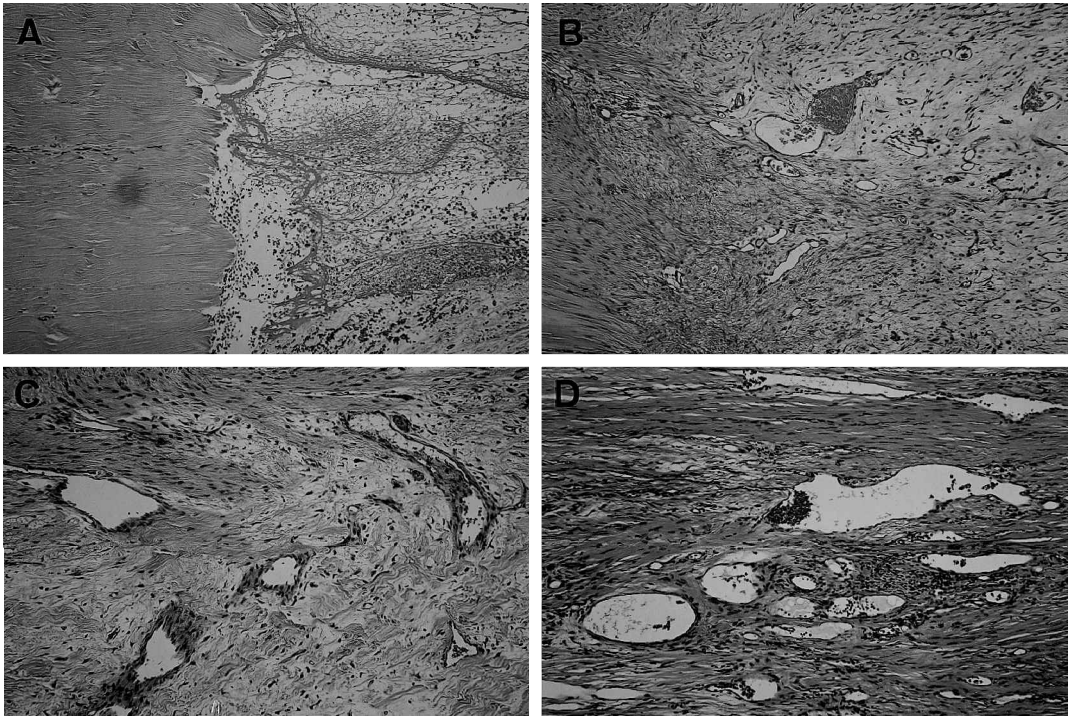


Figure 3. The Achilles tendon. H and E, $\times 100$. A, B, C, and D. Specimens from group B at 1, 2, 3, and 4 weeks, respectively, after surgery (immobilization of both the ankle and knee joints).

return in the healing tendon, just as in the skeletal muscles. Adequate blood supply is a key factor in tendon healing. Accordingly, we believe that when a ruptured Achilles tendon remains static due to immobilization of both the knee and ankle, congestion occurs in the healing tissue, so that tendon regeneration becomes uneven and irregular.

Our study shows that immobilization of the knee and ankle prevents the collagen fiber bundles from aligning parallel to one another along the axis of the tendon. Obviously, a tendon consisting of collagen fiber bundles running in one direction is stronger than one composed of irregularly arranged collagen fibers. Reportedly, tension appears to align fibroblasts in a parallel fashion along the lines of force (Bunting and Eades 1926, Becker and Diegelmann 1984). Accordingly, we believe that an appropriate tension applied on the severed Achilles tendon, due to knee movement may help to arrange the bundles of collagen fibers along the long axis of the tendon. Excessive tension applied on the Achilles tendon may induce elongation of the gap between severed tendon ends, but the present study shows that elongation of the gap following immobilization of only the ankle was not significantly different from that following immobilization of both the knee and ankle joints.

We believe that immobilization of both the knee and ankle is unfavorable for healing the ruptured Achilles tendon, because the static condition of the tendon does not provide adequate blood supply and suitable tension for healing.

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