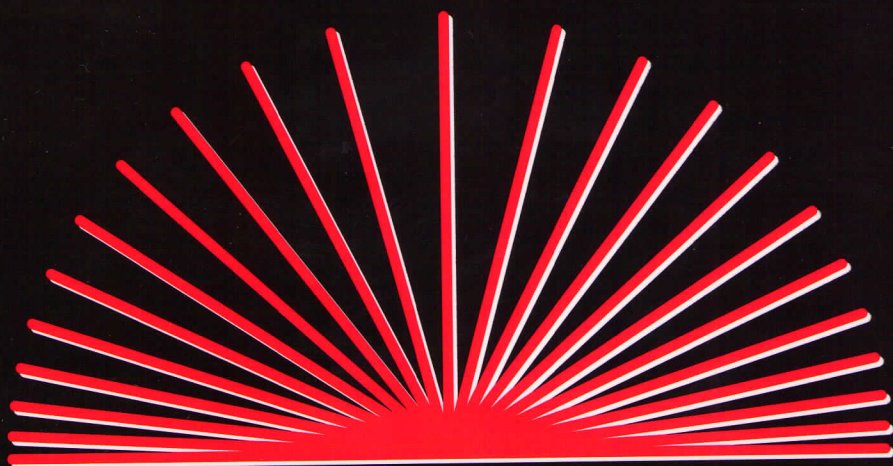

VOLUME 18, NUMBER 4

November 2002

Journal of

APPLIED

BIOMECHANICS



A Modified Cryo-Jaw for In Vitro Biomechanical Testing of Tendons

Federico Morelli, Andrea Ferretti, Fabio Conteduca,
Francesca Nanni, Lucilla Monteleone, and Marco Valente
University of Rome "La Sapienza"

The purpose of this study was to develop a new device, which represents a modification of the Cryo-Jaw described by Riemersma and Schamhardt and modified by Hamner et al., for in vitro biomechanical testing of tendons which allows the lower clamp to move in every direction and thus simulate a pathological dislocation of the knee. Tendons are fixed to the device by freezing the clamped part with dry ice. After fixation of their free ends, the lower clamp was rotated 45°, translated 1 cm, and angled 40° to simulate a knee sprain. Various configurations of bundles were tested: parallel, twisted, and braided. Tests were performed on 10 paired bovine bifurcated digital extensor tendons and 6 paired human hamstring tendons. Grafts were then tested to failure subjected to impulsive load, using a servohydraulic machine. The highest ultimate load recorded for parallel bundles was 4662 ± 565.71 N for bovine bifurcated digital extensor tendons, and 3057 ± 475.44 N for human hamstring tendons. In any case, the tendons ruptured midway, well clear of the frozen part; in no case was slippage of the tendons observed. Thus the device proposed allows one to test what happens to the graft of an ACL reconstructed knee during physiological and pathological movements because it can be easily displaced in every direction.

Key Words: ACL, hamstrings, clamp, dry ice

Introduction

Anterior cruciate ligament (ACL) reconstruction with autologous grafts is a common procedure in orthopaedic surgery. Bone-patellar tendon-bone (BPTB) and doubled gracilis and semitendinosus tendons (DGST) are usually used. In the last

Morelli, Ferretti, and Conteduca are with the Dept. of Orthopaedic Surgery, Hospital S. Andrea; Nanni and Valente are with the Dept. of Chemical and Materials Engineering; and Monteleone is with the Dept. of Mechanics and Aeronautics; Univ. of Rome "La Sapienza," Italy.

A Modified Cryo-Jaw

decade, biomechanical properties of the grafts to optimize the result of ACL reconstruction and force/elongation curve of the grafts to physiological and pathological knee movements can be easily fixed to the testing machine. The collagenous structure of the tendons. Besides, as with DGST, there could be no properties. Soft tissue may be markedly compressive compression. Moreover, there is a device and wet, soft collagenous tissues.

Many researchers have studied the properties (Abrahams, 1967; Benedict, 1967; McCrum, 1976; Viidik, 1967). A clamp (1982), is commonly used. This allowed and damage of the tendons within and (Cryo-Jaw). Recently this device has been used by Hecker, & Hayes (1999) and by MacWilliams in their dynamic knee simulator. From loads with disruption of the tendons from the clamps. All devices used today describes a new device for in vitro biomechanical testing of the lower clamp to move in every direction of the knee.

The two clamps are made of steel and that can be tightened together, between the tendons are 1 mm deep and have a rounded rear portion of these clamps is a central clamp can be displaced laterally and can be connected to a servohydraulic testing machine placed at the top and bottom of the knee.

After definitive revision of the device, 10 paired bovine bifurcated digital extensor tendons. The tendons were secured to the jaws: the four screws of the jaw were driven up to 5 Nm diagonally. Then the tendons were frozen. Various configurations of bundles were tested: parallel, twisted, and braided. The upper grip received the tendons after fixation of the free ends of them. The lower clamp was rotated 45°, translated 1 cm, and the testing machine was able to simulate inertial effects of the clamps. The grafts were tested to failure using a servohydraulic testing machine every 50 ms).

decade, biomechanical properties of these tendons have been widely studied to optimize the result of ACL reconstruction. Knowing the stiffness, pullout strength, and force/elongation curve of the graft is essential to understanding its behavior in physiological and pathological knee movements. Bony ends of tendons, as in BPTB, can be easily fixed to the testing machine in several ways without damaging the collagenous structure of the tendons. If the bony ends are missing on one or both sides, as with DGST, there could be problems when testing the biomechanical properties. Soft tissue may be markedly deformed and rapidly damaged by excessive compression. Moreover, there is very low friction between the material of the device and wet, soft collagenous tissues.

Many researchers have studied different devices so as to avoid these difficulties (Abrahams, 1967; Benedict, Walker, & Harris, 1968; Cohen, Hooley, & McCrum, 1976; Viidik, 1967). A clamp, as described by Riemersa and Schamhardt (1982), is commonly used. This allows excellent grip without major deformation and damage of the tendons within and exterior to the frozen region of the clamp (Cryo-Jaw). Recently this device has been modified by Hamner, Brown, Steiner, Hecker, & Hayes (1999) and by MacWilliams, Wilson, DesJardins, and Chao (1999) in their dynamic knee simulator. Freezing the clamps allows for testing to high loads with disruption of the tendons in the midsubstance without their slipping from the clamps. All devices used today can test tendons only axially. This paper describes a new device for *in vitro* biomechanical testing of tendons which allows the lower clamp to move in every direction, thus simulating a pathological dislocation of the knee.

Methods

The two clamps are made of steel and aluminium. Both have two indented plates that can be tightened together, between which the tendons are placed. The indentations are 1 mm deep and have a rounded top to avoid damage to the tendons. In the rear portion of these clamps is a container in which dry ice is placed. The lower clamp can be displaced laterally and can be rotated and angled. Both clamps can be connected to a servohydraulic testing machine (Instron 5584) by a steel pin placed at the top and bottom of the clamps (Figures 1 and 2).

After definitive revision of the device, preliminary tests were performed on 10 paired bovine bifurcated digital extensor tendons and 6 paired human hamstring tendons. The tendons were secured to the clamp with moderate compression of the jaws: the four screws of the jaws were tightened with a dynamometric screwdriver up to 5 Nm diagonally. Then the jaws and the clamped part of the tendons were frozen. Various configurations of bundles were tested: parallel, twisted, and braided. The upper grip received the axilla of the looped tendons; the lower grip, after fixation of the free ends of them, was rotated 45°, translated 1 cm, and angled 40° to simulate a knee sprain (Figure 3). The two jaws were set at a distance of 9 cm, and the testing machine was adjusted every time to avoid any influence of inertial effects of the clamps. The graft was then subjected to impulsive load (different tests were performed at various speeds: 180, 360, and 540 mm/min) and tested to failure using a servohydraulic machine. Data were sampled at 20 Hz (one every 50 ms).

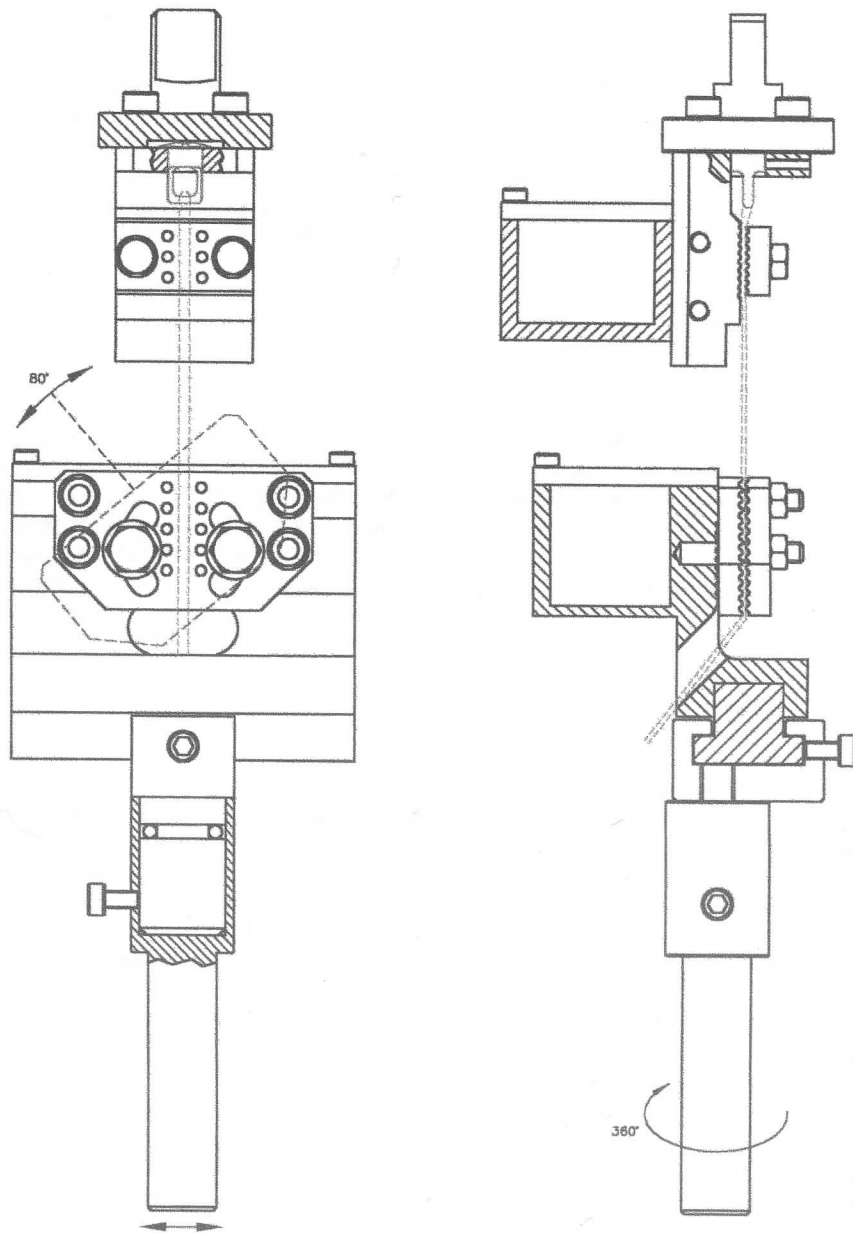


Figure 1 — The modified Cryo-Jaw for biomechanical testing of tendons.

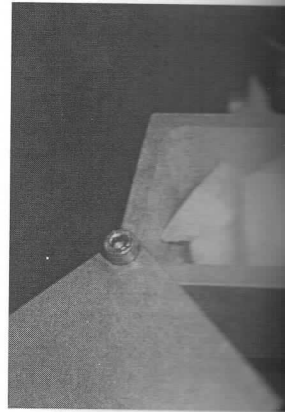
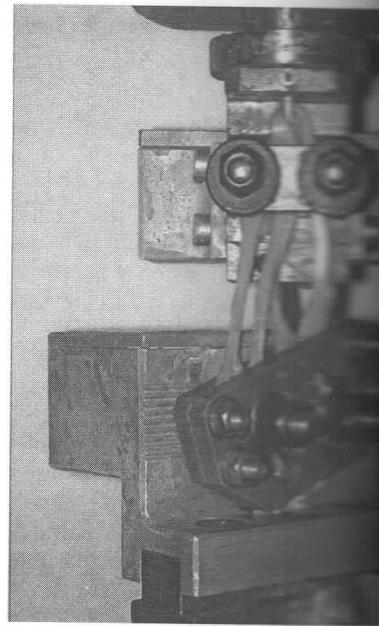


Figure 2 — The posterior container



The highest ultimate load recorded was 4662; mean: 3846.43; SD: 565.71 and 3057 N (range: 1804–3057; mean tendons. The highest stiffness recorded

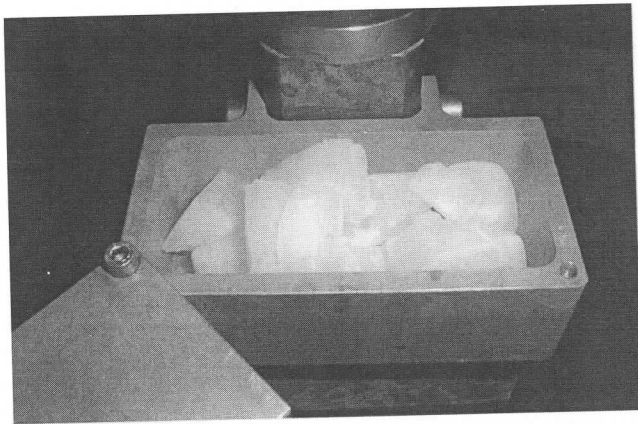


Figure 2 — The posterior container for dry ice on both clamps.

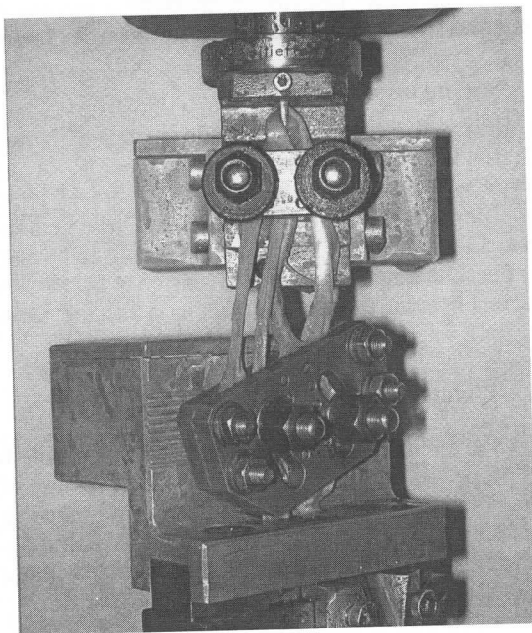


Figure 3 — The lower grip, after freezing the tendons, is rotated 45°, translated 1 cm, and angled 40°, simulating a knee sprain.

Results

The highest ultimate load recorded for parallel bundles was 4662 N (range: 3114–4662; mean: 3846.43; *SD*: 565.71) for bovine bifurcated digital extensor tendons, and 3057 N (range: 1804–3057; mean: 2428.33; *SD*: 475.44) for human hamstring tendons. The highest stiffness recorded was 663 N/mm (range: 376–663; mean:

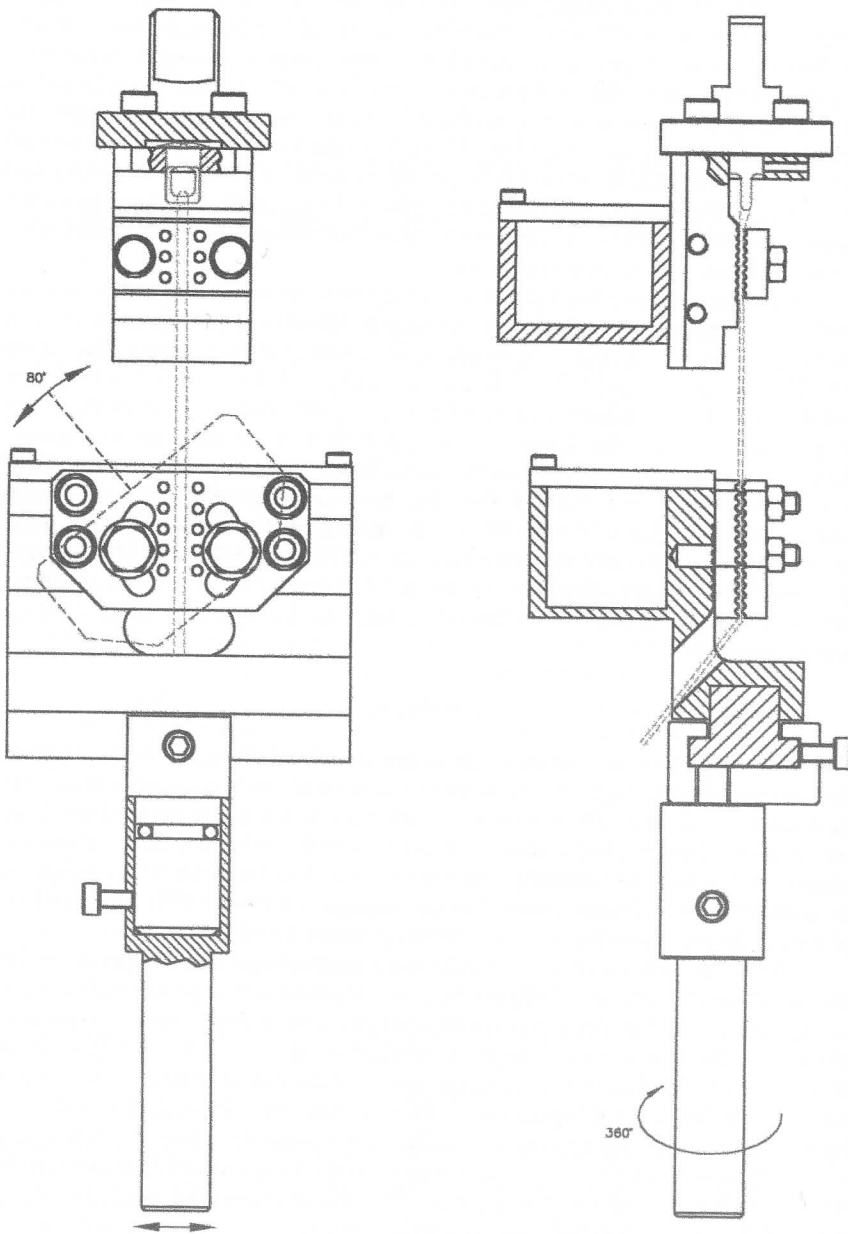
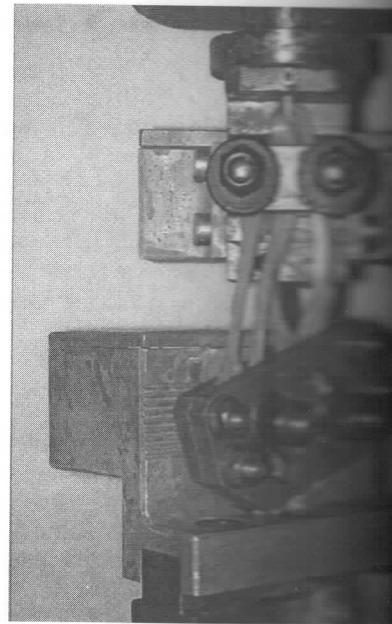


Figure 1 — The modified Cryo-Jaw for biomechanical testing of tendons.



Figure 2 — The posterior container.



The highest ultimate load recorded was 4662; mean: 3846.43; SD: 565.71 and 3057 N (range: 1804–3057) tendons. The highest stiffness recorded

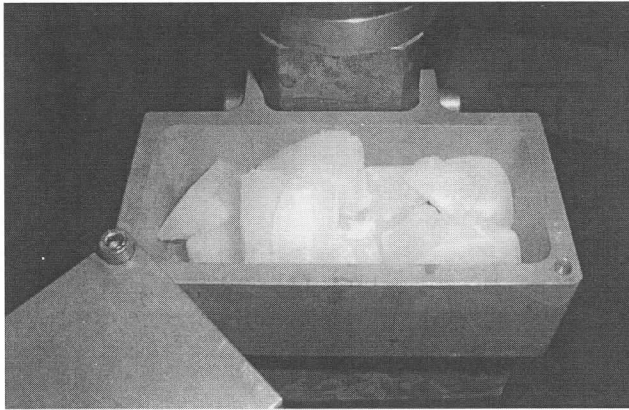


Figure 2 — The posterior container for dry ice on both clamps.

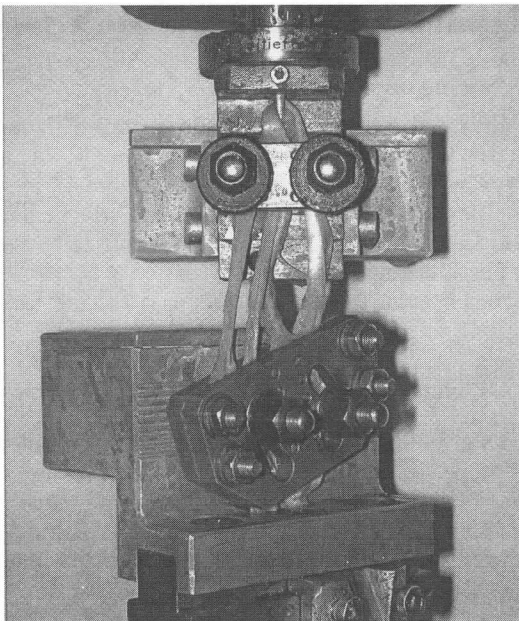


Figure 3 — The lower grip, after freezing the tendons, is rotated 45°, translated 1 cm, and angled 40°, simulating a knee sprain.

Results

The highest ultimate load recorded for parallel bundles was 4662 N (range: 3114–4662; mean: 3846.43; *SD*: 565.71) for bovine bifurcated digital extensor tendons, and 3057 N (range: 1804–3057; mean: 2428.33; *SD*: 475.44) for human hamstring tendons. The highest stiffness recorded was 663 N/mm (range: 376–663; mean:

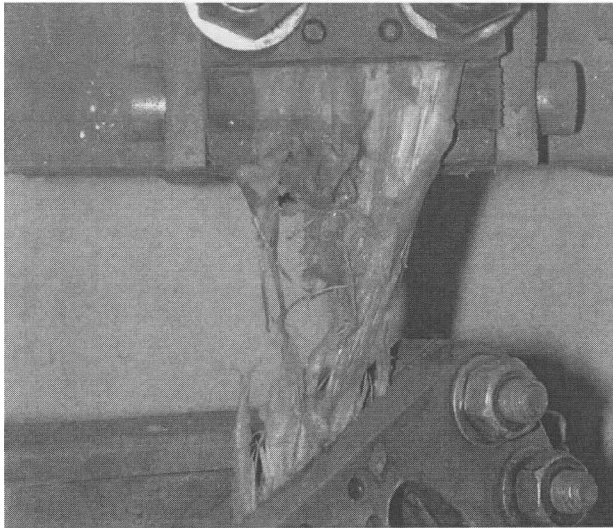


Figure 4 — The rupture of the tendons is in their midsubstance, far from the frozen part.

471.08; *SD*: 113.87) for bovine bifurcated digital extensor tendons, and 480 N/mm (range: 192–480; mean: 310.33; *SD*: 97.39) for human hamstring tendons. In any case, the tendons ruptured midway, well clear of the frozen part (Figure 4). In no case was slippage of the tendons observed. The Cryo-Jaw system has been demonstrated to reach ultimate load up to 13,800 N (with digital flexor tendons of horses) with no slippage. It would not be possible for this tremendous tension to be reached with human grafts, demonstrating that freezing free ends of tendons is a secure, satisfactory, and reproducible manner for fixing wet, soft tissue to testing machines.

Discussion

The wet interface between soft tissue and the clamping device is a major problem when testing the biomechanical properties of the tendons. Excessive pressure of jaws can lead to deformation and/or lesion of the clamped part of the tendons. Moreover, friction between inner collagenous fibers is lower than the friction between outer fibers and the clamp, resulting in undetected slipping of the inner fibers.

Freezing the jaws to the clamped part of the tendons overcomes this problem. Only reduced pressure is applied to the jaws so that the clamped parts of the tendons are only deformed by the indentations of the jaws but are not disrupted by them. Dry ice is applied since the tendons are frozen up to the undeformed zone a few millimeters away from the clamp. Then compression is increased without increasing the deformation of the tendons because they are frozen. At this stage the frozen profiles of the tendons are gripped tightly in the jaws of the clamps. The temperature of the tendon must be carefully monitored, taking care not to freeze the midsubstance. Hamner et al. (1999) could not find any significant difference in

the biomechanical behavior between room temperature.

In the last decade many researches of hamstrings tendons either at room temperature or at low temperature were performed axially. Hamner et al. (1999) tested a gracilis and semitendinosus graft as BPTB graft when tested axially. Moreover, the four bundles are under equal tension, which is different from what happens during physiological loading. In fact, Wallace, Howell, and Hall (1998) showed that the tensions of the four bundles are not equal.

The improvement using our device has been presented for biomechanical testing. The tendons are displaced in every direction to simulate physiological loading and ultimate loads can be measured. The Cryo-Jaw system allows tendons to be tested at the same time in every direction: parallel, twisted, and braided. This is a reproducible manner, which is useful during physiological and pathological loading.

- Abrahams, M. (1967). Mechanical behavior of tendons. *Journal of Biomechanical Engineering*, 5, 433–443.
- Benedict, J.V., Walker, L.B., & Harris, E.H. (1967). The strength of unembalmed human tendons. *Journal of Biomechanics*, 1, 1–10.
- Cohen, R.E., Hooley, C.J., & McCrossin, M. (1980). The use of tissue. *Journal of Biomechanics*, 13, 101–106.
- Hamner, D.L., Brown, C.H., Steiner, W.E., & Goss, J.D. (1999). Tendon grafts for reconstruction: A comparative evaluation of the use of multiple tendons. *Journal of Orthopaedic and Joint Surgery [Am.]*, 11A, 544–549.
- MacWilliams, B.A., Wilson, D.R., & Goss, J.D. (1998). Cocontraction reduces internal moment load in weight-bearing tendons. *Journal of Biomechanics*, 31, 793–799.
- Nicklin, S., Waller, C., Walker, P., & Goss, J.D. (1998). The properties of braided tendon grafts. *Journal of Biomechanics*, 31, 793–799.
- Riemersma, D.J., & Schamhardt, H.C. (1998). The rheology studies of horse digital tendons. *Journal of Biomechanics*, 31, 620–626.
- Viidik, A. (1967). Experimental evaluation of tendon grafts. *Journal of Biomedical Engineering*, 2, 94–100.
- Wallace, M.P., Howell, S.M., & Hall, M.J. (1998). The hamstring graft as a replacement for the anterior cruciate ligament. *Journal of Paediatric Research*, 15, 534–540.

the biomechanical behavior between tendons tested at 13 °C and those tested at room temperature.

In the last decade many researchers have studied the biomechanical properties of hamstrings tendons either at impulsive or cyclic loading. But all tests were performed axially. Hamner et al. (1999) demonstrated that the four parallel bundles of a gracilis and semitendinosus graft are much stronger and stiffer than ACL and BPTB graft when tested axially. Moreover, ultimate load and stiffness increases if the four bundles are under equal tension. Unfortunately, this situation is very different from what happens during physiological and pathological knee movements. In fact, Wallace, Howell, and Hull (1997) have demonstrated that the peak tensions of the four bundles are not equal during passive motion.

The improvement using our device is that, unlike all other similar devices that have been presented for biomechanical testing of tendons, it can be easily displaced in every direction to simulate traumatic dislocations of the knee. Cyclic loading and ultimate loads can be measured. The device can be easily secured to a servohydraulic materials testing machine. This new device allows two doubled tendons to be tested at the same time. Almost every configuration of the bundles is possible: parallel, twisted, and braided. We believe the device proposed allows one to test, in a reproducible manner, what happens in an ACL-reconstructed knee during physiological and pathological movements.

References

- Abrahams, M. (1967). Mechanical behaviour of tendon in vitro. *Medical & Biological Engineering*, **5**, 433-443.
- Benedict, J.V., Walker, L.B., & Harris, E.H. (1968). Stress-strain characteristics and tensile strength of unembalmed human tendon. *Journal of Biomechanics*, **1**, 53-63.
- Cohen, R.E., Hooley, C.J., & McCrum N.G. (1976). Visco-elastic creep of collagenous tissue. *Journal of Biomechanics*, **9**, 175-184.
- Hamner, D.L., Brown, C.H., Steiner, M.E., Hecker, A.T., & Hayes W.C. (1999). Hamstring tendon grafts for reconstruction of the anterior cruciate ligament: Biomechanical evaluation of the use of multiple strands and tensioning technique. *Journal of Bone and Joint Surgery [Am.]*, **81A**, 549-557.
- MacWilliams, B.A., Wilson, D.R., DesJardins, D.J., & Chao, Y.S. (1999). Hamstrings cocontraction reduces internal rotation, anterior translation, and anterior cruciate ligament load in weight-bearing flexion. *Journal of Orthopaedic Research*, **17**, 817-822.
- Nicklin, S., Waller, C., Walker, P., Chung, W.K., & Walsh, W.R. (2000). In vitro structural properties of braided tendon grafts. *American Journal of Sports Medicine*, **28**, 790-793.
- Riemersa, D.J., & Schamhardt, H.C. (1982). The Cryo-Jaw, a clamp designed for in vitro rheology studies of horse digital flexor tendons. *Journal of Biomechanics*, **15**, 619-620.
- Viidik, A. (1967). Experimental evaluation of the tensile strength of isolated rabbit tendons. *Biomedical Engineering*, **2**, 64-67.
- Wallace, M.P., Howell, S.M., & Hull, M.L. (1997). In vivo tensile behavior of a four-bundle hamstring graft as a replacement for the anterior cruciate ligament. *Journal of Orthopaedic Research*, **15**, 539-545.