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EFFECTIVENESS OF EXERCISE AND JOINT-MOBILISATION ON THE BIOMECHANICAL PROPERTIES OF THE IMMOBILISED RABBIT'S KNEE

BY

BARBARA E. ROBINSON

A THESIS

SUBMITTED TO THE FACULTY OF GRADUATE STUDIES AND RESEARCH

IN PARTIAL FULLFILLMENT OF THE REQUIREMENTS FOR THE DEGREE

OF MASTER OF SCIENCE

DEPARTMENT OF PHYSICAL THERAPY

EDMONTON, ALBERTA

SPRING, 1988

ISBN 0-315-42831-7

THE UNIVERSITY OF ALBERTA

RELEASE FORM

NAME OF AUTHOR: Barbara E. Robinson

TITLE OF THESIS: Effectiveness of Exercise and Joint-Mobilisation on the Biomechanical Properties of the Immobilised Rabbit's Knee

DEGREE: Master of Science

YEAR THIS DEGREE GRANTED: 1988

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The undersigned certify that they have read, and recommend to the Faculty of Graduate Studies and Research for acceptance, a thesis entitled Effectiveness of Exercise and Joint-Mobilisation on the Biomechanical Properties of the Immobilised Rabbit's Knee submitted by Barbara E. Robinson in partial fulfillment of the requirements for the degree of Master of Science.

(Supervisor)

Date: April 25/ 1988

DEDICATION

To Daijf, my love
To our dreams

ABSTRACT

This study analyzed the effects of specific joint-mobilisation and exercise on the amount of stiffness, in a previously immobilised joint. Treated and control knees were compared in terms of amount of torque and areas of hysteresis (utilizing torque-angular displacement curves of a flexion-extension cycle for an immobilised rabbit's knee):

i) Group 1 immediately following immobilisation (8 weeks),

ii) Group 2 following remobilisation in the form of unlimited free active exercise (1 week), and

iii) Group 3 following remobilisation in the form of unlimited free active exercise, in conjunction with specific joint-mobilisation (1 week).

The left hind limb of the rabbit was immobilised in a position of full knee flexion by means of a surgically inserted Steinmann pin. All biomechanical testing was performed on an arthrograph.

Statistically significant results were found for areas of hysteresis for treated knees (between Groups 2, and 3), for first cycle stiffness slopes for treated knees (between Groups 1 and 2, and also between Groups 1 and 3), for angular displacement (between Groups 1 and 2, and also between Groups 1 and 3) and for the ratio of vertical to angular displacement (between Groups 1 and 3). Results for Group 3 revealed the smallest area of hysteresis and the least joint stiffness (slope). The beneficial effects of specific joint-mobilisation were decidedly apparent in 1st cycle areas of hysteresis.

ACKNOWLEDGEMENTS

I would like to thank my advisor, Dr. David Magee for his generous support, encouragement, and patience throughout the entire thesis process.

I would also like to thank the members of my committee: Dr. David Budney. He initiated the final arthrograph design and engaged in countless hours of insightful discussion of my research findings and their possible significance. Dr. David Reid. He helped me to establish the initial research design, helped interpret roentgenograms and arthrographic data, and facilitated the histological preparations. An adroit crafter of English, freely giving of expertise, time and encouragement, he helped immeasurably in giving me a realistic view of research and simply finishing. Dr. Jean Wessel. She made statistics more real and comprehensible, possibly even enjoyable. Through the writing of the thesis she was a voice of clarity and organization, wading through revisions revised, and ringing positive throughout.

As well, I am indebted to Dr. David Secord. He was an original committee member, always readily available with his expertise, inspiration, and tremendous patience. He was exceedingly helpful in my gaining understanding of animals and surgery.

I would like to express thanks for the financial assistance I received from the M.S.I. Foundation, Department of Physical Therapy — University of Alberta, and the Canadian Physiotherapy Association, Edmonton District.

In addition I would like to recognize and thank:

- Freddy Kaltenborn and Olaf Evjenth for nurturing my skills in joint-mobilisation
- Dr. Shrawan Kumar for contributing to my understanding of tissue biomechanics, a vital prerequisite to the theoretical background to my research
- Guy Hervieux for giving generously of his time and expertise in the arthrograph design, construction and operation, and helping me to more fully understand biomechanical engineering

- the efforts of the 4th year biomechanical engineering design class, 1986 in solving the problems of the initial arthrograph design
- the surgical technicians in Small Animal Surgery: Ted Germaine for his constant support and guidance; Peter for assisting during the surgeries of the pilot studies; and Alvin for assisting in surgery during the final study
- the Biomechancial Engineering technicians, Al Muir and Max for their exacting work in construction of the final arthrograph, and for their patience
- Margaret Wright for valuable help in solving the problem of fracturing
- Doug Booth for instruction in and use of the digitizer
- Rafe for his superb illustrations
- Valeska Phipps for her constant support and giving
- my parents, Dr. and Mrs. John and Jessica Robinson for their love and support
- Mr. and Mrs. Ernie and Gladys Krauss for their generous giving in whatever ways they could - typing, proofing, meals, more, and hope
- Daijf for making it all possible.

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I. INTRODUCTION

1.1 Background to the Problem

During immobilisation of a joint, there are changes both in the periarticular soft tissues surrounding the joint and in the articular cartilage with its underlying subchondral bone.[3, 6, 35, 76, 264] Over time these changes result in joint contracture involving a stiffening of the joint.[176]

The fibrofatty tissue proliferation within the joint space forms strong adhesions between non-contacting articular surfaces and between synovial folds of the joint capsule.[76, 87] Regions with contacting articular cartilage develop changes consistent with osteoarthritis, with localized areas of compression and eventual erosion of superficial cartilaginous layers. The underlying subchondral bone becomes either atrophic or cystic.[68]

Furthermore, during immobilisation of a joint, organized extra-articular structures, such as ligaments and tendons, lose the orientation of their collagenous fibres and as a result have greatly diminished strength.[3] Weakening also occurs at ligamentous insertion sites on the bone, due to absorption of bone at these sites.[14, 87, 131]

In conjunction with this overall structural weakening, biochemical alterations in loss of both water and glycosaminoglycans (GAGS) impairs the normal gliding that occurs between collagenous fibres.[6, 10, 14, 87] It has been noted that with experimentally-induced contractures in animals, there occur biochemical and biomechanical changes which parallel observations made of human joint contractures.[87, 90, 91]

The reactions of these stress-deprived (immobilised) tissues to a period of remobilisation consisting of exercise show that motion, whether active or passive, partially reverses these changes.[68, 76, 158, 166, 176] With repeated motion, the tissues appear not only to become stretched, but lubricated as well.[76] One of the benefits of exercise for normal joint soft tissue is a gradual increase in biomechanical strength — over the long term with consistent exercise.[57, 87, 217, 220, 253]

There are numerous theories regarding the biomechanical effects of specific joint-mobilisation, but with little scientific evidence in support.[40, 64, 71, 172, 173] Nonetheless, there is much interest in joint-mobilisation as a means of dramatically improving joint range of motion. However, treatment decisions are based largely on clinical experience and anecdotal evidence.

Despite overwhelming evidence that joint-immobilisation is deleterious to articular and periarticular tissues from anatomical, physiological, and biomechanical viewpoints,[14, 220] the practice of joint-immobilisation is often necessary during fracture healing or after surgical repair. There is now sufficient evidence for clinicians to utilize motion, active or passive, after immobilisation to improve, enhance, and restore normal functioning of tendons, ligaments, joints, and muscles.[220]

However, many questions remain unresolved, such as the eventual extent of recovery possible after immobilisation and the influence of post-immobilisation activities in type, intensity, duration, and overall time-frame.[3, 14, 220] Even the effect that duration of immobilisation has on the potential ability of tissues to return to normal functioning remains unknown.[8, 68, 75, 87, 91, 162] Similarly, the effects of joint-mobilisation, with or without an active exercise programme, remain key unresolved questions.

It was against this background of basic, broad, and unanswered questions about immobilisation that this thesis investigated joint-mobilisation, widely acknowledged in clinical efficacy, yet at present lacking a strong, integrated scientific basis.

1.2 Objectives

The objective was to study, for a previously immobilised joint, the effect of specific joint-mobilisation on the resistance offered to passive movement.

The present investigation compared treated and control knees by measuring torque and areas of hysteresis (utilizing torque-angular displacement curves of a flexion-extension cycle for an immobilised rabbit's knee):

a) immediately following immobilisation,

b) following remobilisation in the form of unlimited free active exercise, and

c) following remobilisation in the form of unlimited free active exercise, in conjunction with specific joint-mobilisation.

1.3 Primary Research Hypotheses

H_1: Areas of hysteresis for control knees are essentially the same for each group and for each of the three cycles.

H_2: Areas of hysteresis for treated knees vary from group to group and cycle to cycle. Regardless of cycle, the areas of the immobilised-only group (Group 1) are largest, with the areas of the exercised joint-mobilised group (Group 3) the smallest. The areas of the exercised group (Group 2) are slightly larger than those of Group 3. For all groups, the 1st cycle area is the largest, with each subsequent cycle area being much smaller.

H_3: The hysteresis area ratio of 1st cycle to 2nd cycle (treated knees as compared to control knees) is greatest for Group 1 and smallest for Group 3. The area ratio of Group 2 is slightly greater than that of Group 3.

H_4: Torque or 'stiffness' is minimal for control knees and essentially the same for each treatment group.

H_5: **End-range stiffness slopes (torque) for treated knees demonstrate greatest stiffness in the 1st cycle of all groups and are much lower starting with the 2nd cycle of all groups. Torque is greatest in Group 1 and least in Group 3, with Group 2 torque slightly more than that of Group 3.**

H_6: First cycle stiffness slopes (torque) of the treated knees are greatest for Group 1 and least for Group 3, with Group 2 slightly more than Group 3.

1.4 Operational Definitions

1 Joint stiffness:

Joint stiffness is defined as "the resistance to passive motion at a joint throughout the normal range of motion in the usual functional plane." [215]

2. Range of motion:

The amount of motion or movement available at a specific joint is conventionally termed range of motion (ROM). Limb movement, expressed in degrees, was written as 48° - 98° - 48° ROM (flexion-extension-flexion ROM). With 180° taken as the position of full knee extension, the knee was first extended from 48°-98° and then flexed from 98°-48°.

3 Hysteresis loop:

The changing angles and forces required during cycling of the rabbit's knee through flexion-extension-flexion ROM on the arthrograph created a torque-angular displacement curve, or hysteresis loop. Angular displacement was recorded on the X axis (horizontal) and torque on the Y axis (vertical) (Figure I-1).

4. Measurements from the hysteresis loop:

a) Area

The area of hysteresis was directly related to the amount of stiffness present in a joint.[254] The larger the enclosed area, the greater was the stiffness present within the joint (Figure I-1). The area measured represented the amount of energy required to cycle the limb through the entire flexion-extension-flexion passive ROM.

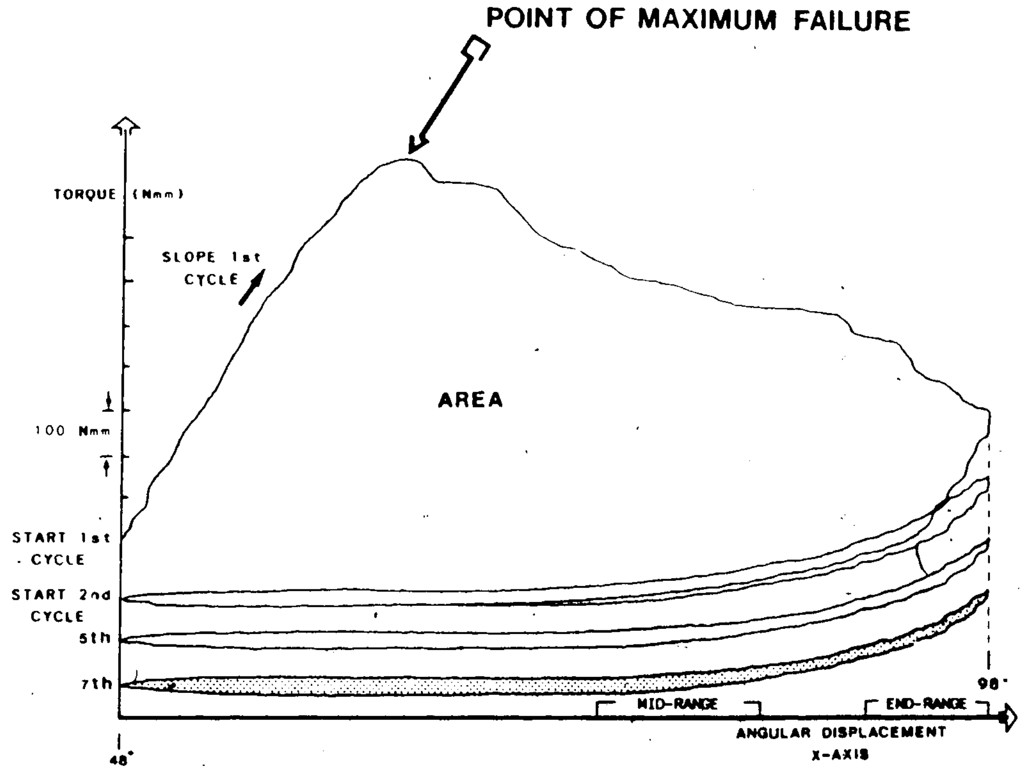


FIGURE I-1 Hysteresis Loops for 1st, 2nd, 5th, and 7th Cycles in a Flexion-extension-flexion Cycle (48°-98°-48°) (Rabbit #33 Immob-only). Angular displacement (50° ROM) is recorded on the X axis and torque on the Y axis. The larger the enclosed area of hysteresis, the greater is the stiffness present within the knee joint. The steeper the 1st cycle slope, the greater is the stiffness. The point of maximum failure plus mid-range (73°-85°) and end-range (92°-98°) regions are indicated.

b) Stiffness

i) torque (moment of force)

The magnitude of torque was expressed as the product of force (in Newtons) and moment arm (in millimeters). Therefore, the unit for measurement of torque was Newton millimeters (Nmm).

The moment arm was expressed as the perpendicular distance from the line of force application to the centre of rotation. It was around this axis that the torque or moment acted. The axis point of the moving arthrograph arm coincided with the axis point of the knee joint.

ii) slope

The slope of the knee extension portion of the torque-angular displacement curve represented the stiffness or resistance to passive knee motion (Figure I-1). In the context of this study, first cycle slope or stiffness was torque (Nmm vertical displacement) divided by angular displacement (degrees),[53] or in simpler terms, rise over run. The steeper the slope, the greater was the stiffness.

iii) mid-range and end-range stiffness

From the starting position of 48° knee flexion and cycling to the end of the range at 98° knee extension, mid-range was considered to be 73° - 85° and end-range 92° - 98°. With a constant angular displacement for both mid-range and end-range stiffness, vertical displacements, as representing stiffness in those particular ranges, were compared in millimeters only (Figure I-1). To convert vertical displacement from millimeters to Nmm, the millimeter value was multiplied by 7.88N. Increased vertical displacement with a constant angular displacement indicated increased stiffness.

iv) ratio of vertical displacement to angular displacement

At the first sign of ligamentous failure on the hysteresis loop (Figure I-1), vertical displacement (mm) and angular displacement (mm) were compared as rise over run for knee extension. The higher the ratio, the more stiffness was present. To convert angular displacement from millimeters to degrees, the millimeter value was multiplied by 0.198°/mm. To convert vertical displacement from millimeters to Nmm, the millimeter value was multiplied by 7.88N.

5. *Joint-immobilisation:*

Joint-immobilisation consisted of a period of eight weeks of internal fixation of the rabbit's left knee in a position of full flexion. There was no restriction of active cage activity or weight bearing within the confines of the fixation.

6. *Remobilisation:*

Remobilisation consisted of a period of one week of free active exercise within the rabbit's pen. The activities of the rabbit were not controlled in any way and weight bearing, as tolerated, was permitted. Remobilisation did not in any way imply aerobic conditioning.

7. *Treatment groups:*

Group 1: Immobilisation-only (Immob-only).

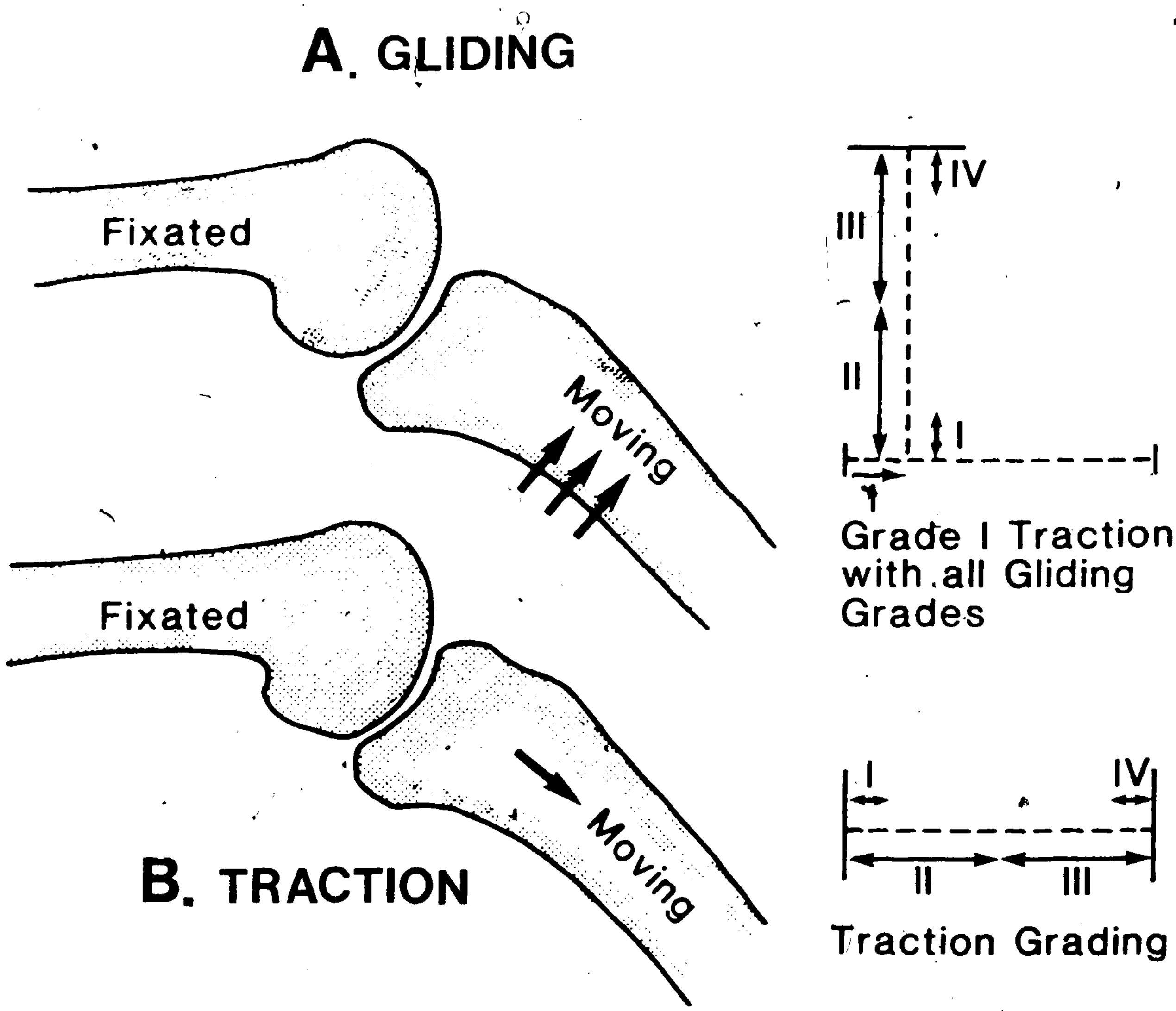
Group 2: Immobilisation followed by a period of remobilisation exercise (Ex).

Group 3: Immobilisation followed by a period of remobilisation exercise in conjunction with specific joint-mobilisation treatments (Ex Jt-Mob).

8. *First, second, fifth, and seventh* cycles: The loading and unloading hysteresis loops through flexion and extension were written as 1st, 2nd, 5th, and 7th cycles.

9. *Joint-mobilisation:*

Joint-mobilisation referred to both passive osteokinematic and passive arthrokinematic movements. A grading system as described by Maitland was utilized: Grades 1 - 4.[141] Graded arthrokinematic movements applied to traction and gliding procedures (Figure I-2). Traction involved a separation of the joint surfaces. Joint gliding between two joint surfaces occurred when the "same point on one surface came into contact with new points on another surface."[117] Gliding to increase knee extension produced an anterior gliding of the tibia in relation to the stationary femur. The starting or resting position corresponded to that position where i) joint surfaces were the least congruent, ii) joint capsule and ligaments exhibited the least tension, and iii) the joint surfaces were most easily separated by traction.[140]



Arthrokinematic Grading

FIGURE I-2 Arthrokinematic Graded Movements (Traction and Gliding). Both traction and gliding manoeuvres can be performed through four grades as illustrated.

A. Gliding: The femur is fixated while the tibia is glided dorsally to improve knee extension.

B. Traction: The femur is fixated while a traction force is exerted along the length of the tibia.

From this initial resting position (almost full knee flexion in the rabbit), the grading system was set up relative to the pathological amplitude of available traction or gliding at the joint.[117] With the joint remaining in the resting position, traction (as a separation of joint surfaces) was performed to the end of the available range. This total available joint range was divided into i) two small amplitude movements, at the beginning (Grade 1) and at the end (Grade 4) of the available range, and ii) two large amplitude movements from the beginning to approximately the middle of the range (Grade 2) and from approximately the middle of the range to the end of the range (Grade 3). With the joint remaining in the resting position, anterior tibial gliding was performed throughout the available range while maintaining a Grade 1 traction.[117] The total available anterior gliding range was also subdivided into Grades 1 - 4.

A passive osteokinematic movement of knee extension from the fully flexed knee position to the limit of knee extension (imposed by the contracture) was also subdivided into four grades (Figure I-3). Both the osteokinematic and arthrokinematic movements (Grades 3 and 4) were used as means of joint-mobilisation to restore the loss in extension of the rabbit's knee.

10. Regions of tensile loading on the load-elongation curve:

Butler et al[45] classified the tensile loading of a ligament into regions on the load-elongation curve (Figure I-4). The initial toe region (Region I) was followed by a fairly linear region (Region II) from which 1st cycle slopes were measured. It was at the end of this linear region that the first evidence of ligamentous failure occurred (point of linear load). Total ligamentous failure occurred at Region IV, following maximum load at failure (Region III). Ratios of vertical to angular displacement were performed at the point of linear load, if obvious, and if not so obvious at the point of maximum load at failure.

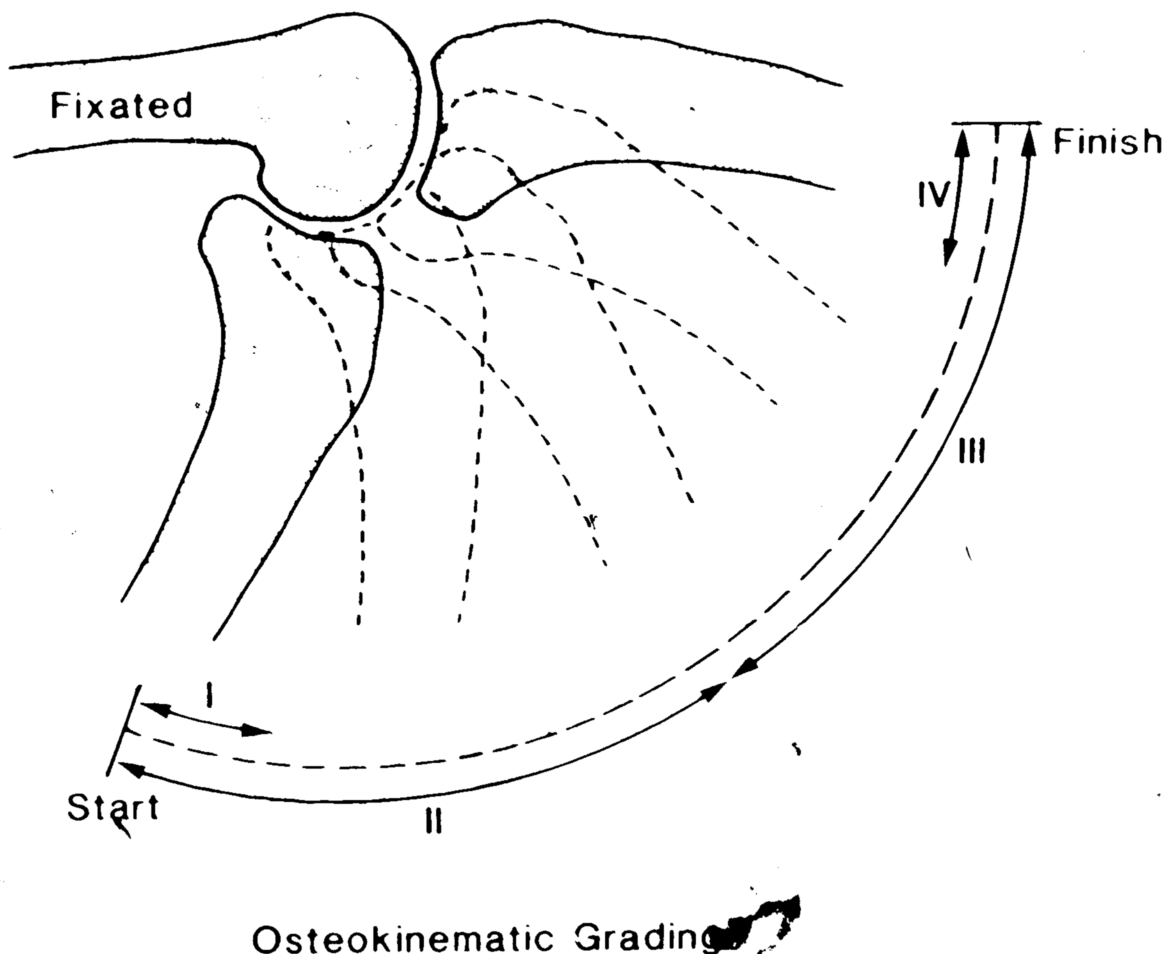


FIGURE I-3 Osteokinematic Graded Movements. The femur remains fixated while the tibia moves from a starting position of near full knee flexion to a position of full knee extension. Graded movements (Grade I-IV) are indicated for this arc of motion.

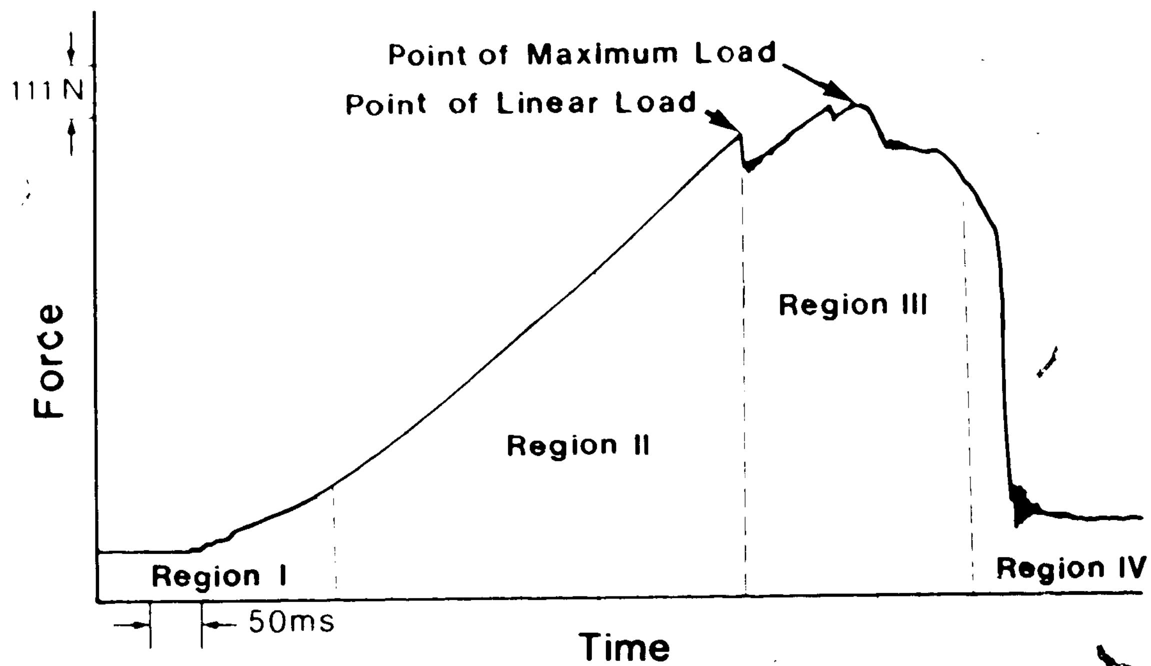


FIGURE I-4 Force-time (Load-elongation) Curve for a Femur-Anterior Cruciate Ligament-Tibia Specimen of a Rhesus Monkey. The initial concave toe region (Region I) is followed by a fairly linear region (Region II) up to the point of linear load which is the first sign of ligamentous failure. Series of sequential failures occur in Region III, with complete failure occurring in Region IV, shortly after the point of maximum load. (Adapted from Butler et al.[45]).

1.5 Delimitations

1. This study was limited to female New Zealand white rabbits weighing from 2.4 to 3.2 kg.

2. The choice of 33 rabbit subjects was intentional to allow for the high attrition rate present with long term studies utilizing rabbits. Groups of five or six rabbits are commonly considered acceptable by researchers (Secord DC: personal communication, 1984).

3. The rabbits were individually housed in pens having overall inside dimensions of 50 cm x 50 cm x 50 cm and a concrete floor covered by wood shavings and sawdust.

4. The investigator performed virtually all surgery and all testing and treatment procedures. The surgical technician assisted by preparing for surgery (autoclaving instruments, anaesthesizing and shaving the rabbit), and during surgery (stabilizing the hind limb, tightening and cutting the Steinmann pin).

5. The same surgical and X-ray personnel assisted throughout the entire study.

6. The same surgical equipment was used from operation to operation.

7. The immobilisation procedure consisted of a Steinmann pin insertion through both tibia and femur, and the use of stabilizing nuts at both ends of the pin.

8. The chosen size of Steinmann pin, 2.4 mm diameter, was consistent with that selected by other researchers[5, 6, 9, 10, 11, 13, 14, 17, 253, 254] and consistent with the size of bone in the chosen weight range.

9. To curb the possibility of infection, long-acting penicillin was administered after each surgery (Secord DC: personal communication, 1984).

10. To produce minimal tissue trauma and bleeding, to promote faster healing, and to shorten the second anaesthetic period, the Steinmann pin was cut instead of removed.

11. The immobilisation period chosen was eight weeks, consistent with that of previous researchers who used either eight or nine weeks.[6, 10, 13, 254]

12. The remobilisation time period chosen was one week, consistent with that of previous researchers who used time periods ranging from one to ten weeks.[11] A one-week period was also chosen to minimize the known high attrition rate in studies utilizing rabbits as subjects (Secord DC personal communication, 1985).

13. The use of the left hind limb as the immobilised knee, and the right hind limb in each rabbit as its own control knee has been described by previous researchers.[5, 6, 9, 10, 11, 13, 14, [illegible], 253, 254]

14. While one excised hind limb was tested, the other remained at room temperature wrapped in saline-moistened towelling to prevent drying of the tissues.[48, 234]

15. The arthrograph was constructed according to the descriptions of Wright and Johns[108, 259, 260] plus Woo et al[254] and subsequently modified according to Budney (personal communication, 1985). A motor speed of 0.2 cycles per second (cps) or 20°/sec matched that chosen by other researchers. It was hoped that with the use of an arthrograph similar to that described by Woo et al,[254] previous research findings would be confirmed and the new dimension of joint-mobilisation added.

1.6 Limitations

1. During immobilisation and treatment periods, the rabbits were housed at Ellerslie Animal Centre. Transport to the main campus was necessary for testing and surgical procedures.

2. The high attrition rate of subjects could only be controlled to a limited extent. Some influencing but only partially-controlled factors were environmental noise levels, correctness of handling technique, and the seemingly inherent vulnerability of rabbits in experimental situations.

3. The amount of periosteal irritation caused by the Steinmann pin in the tibia and femur could not be controlled.

4 Accuracy of the hysteresis loops, which recorded the flexion-extension-flexion cycling of the rabbit's knee, was limited by the design of the constructed arthrograph. Inaccuracies occurred when the axis point of the moving arthrograph arm remained fixed, while the axis point of the knee joint varied throughout the ROM.

5 The velocity of the arc of ROM was dependent on the reliability of the DC power supply.

6 In addition to uncontrollable factors, consistency in surgical, treatment, and testing sessions was limited by the ability of the investigator.

II. LITERATURE REVIEW

2.1 Introduction

The following review provides background information to answer the questions under study relating to the effectiveness of exercise and joint-mobilisation on the biomechanical properties of a previously immobilised rabbit's knee.

An examination of normal ligamentous structural properties leads to a closer examination of the normal biomechanical properties of ligaments and joint capsule. The alteration in these normal biomechanical responses as a result of stress-deprivation (immobilisation),[8] stress-enhancement (exercise),[8] and remobilisation (exercise with or without joint-mobilisation following a period of immobilisation) is presented and related to corresponding biochemical changes.

Treatment and examination of joint stiffness is analyzed from objective and subjective viewpoints. The chapter concludes with the objective measurement and characterization of joint stiffness by means of an arthrograph.

2.2 Ligamentous Structural Properties

The connective tissues of the body, such as connective tissue proper, cartilage, and bone, are classified according to the proportional distribution of their elements — cells and intercellular substances, fibrous and amorphous. The fibrous intercellular substances include collagenous, reticular, and elastic fibres.[132]

The ground substance, an amorphous viscous gel, acts as a supporting structure for the tissues as well as a medium for the transport of tissue fluid containing nutrients and waste products (to diffuse between the cells and capillaries). Formed by connective tissue cells (fibroblasts), the ground substance contains protein, glycoproteins, glycosaminoglycans (GAGS), carbohydrates, lipids, and water. The glycoproteins are polysaccharide-protein complexes, while GAGS are polysaccharides with one or more amino sugar moieties. The GAGS, largely responsible for the viscosity of the ground substance, consist of hyaluronic acid, chondroitin-4-sulfate, chondroitin-6-sulfate, keratan sulfate, dermatan sulfate, and heparan sulfate.[37, 238] Keratan sulfate contains the sugar residues, glucosamine and galactose, while the remainder contain hexosamine and hexuronic acid.[183, 238]

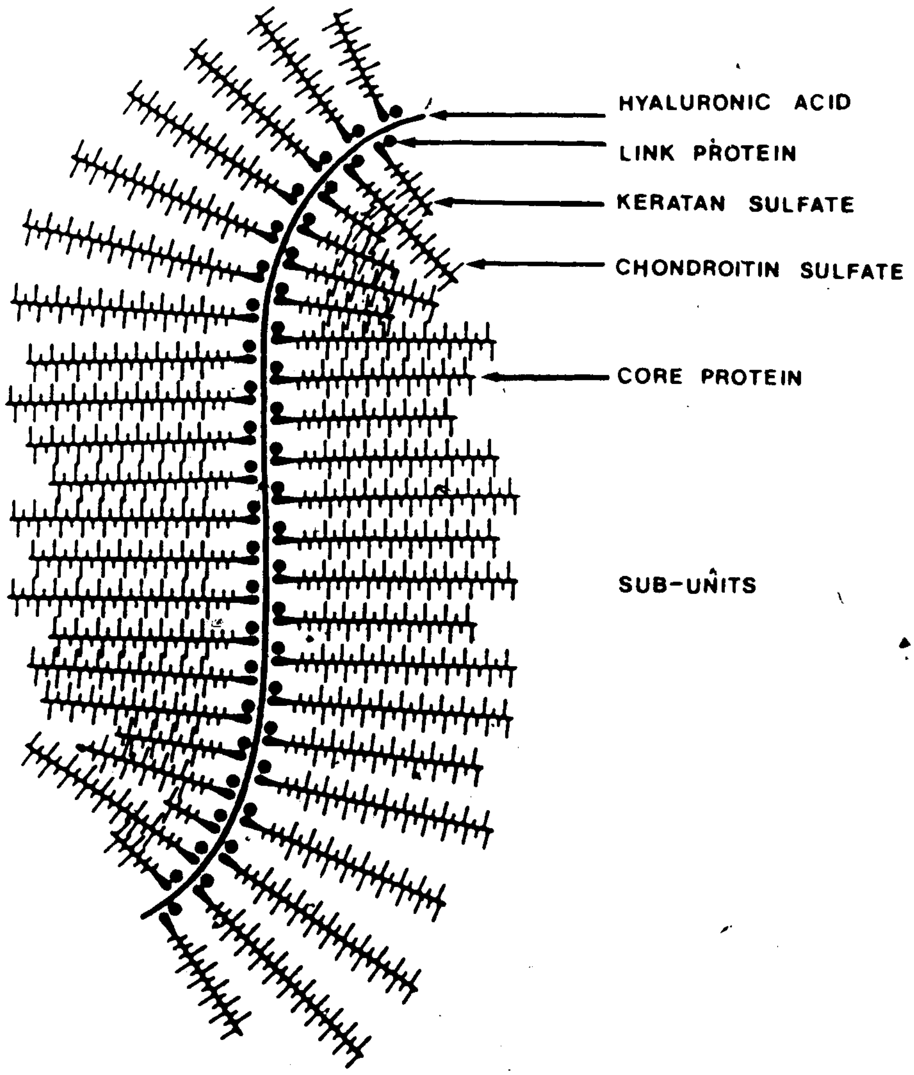
Most GAGS appear in tissues in the form of proteoglycan sub-units ("units of disaccharides coupled as linear polymers onto protein 'cores' ")[238] (Figure II-1). Proteoglycan aggregates consist of proteoglycan sub-units, hyaluronic acid, and link protein.[183] Hyaluronic acid provides the "filamentous backbone" for the attachment of proteoglycan sub-units and link protein to form proteoglycan aggregates.[183]

The proteoglycan sub-unit consists of chondroitin sulfate and keratan sulfate chains attached by means of linkages to a central core protein. The number of sulfate chains present in a proteoglycan sub-unit is determined by the length of the core protein.[183] Both chondroitin sulfate and keratan sulfate "contain closely spaced negatively-charged groups distributed along their polysaccharide chains."[183] Due to the 'repelling forces' of these negatively-charged groups, the GAG chains assume a 'stiffly extended' space-occupying configuration.[183]

Collagenous fibres are present in all the different types of connective tissue. The fibres within a bundle, held together by mucoprotein, are either loosely or densely packed, depending upon their location. The normally 'wavy' appearance on scanning microscopy straightens when the connective tissues are under tension.[132]

Within dense connective tissue, there is a close packing of the fibres, fewer cells than in the loose connective tissues, and less amorphous ground substance. Dense *irregular* connective tissue with its interwoven, randomly oriented fibre bundles is present in areas where multi-directional tensions are exerted, such as in joint capsules, tendon sheaths, periosteum, and fasciae. On the other hand, dense *regular* connective tissue with its parallel arrangement of fibre bundles is present in structures subject to uni-directional tensile forces, such as tendons, ligaments, and aponeuroses.

PROTEOGLYCAN AGGREGATE



PROTEOGLYCAN SUB-UNIT

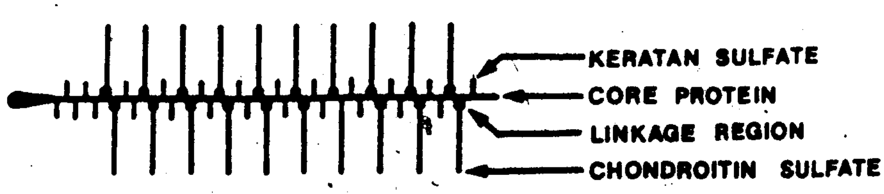


FIGURE II-1. Proteoglycan Aggregate(A) and Sub-unit(B). Proteoglycan aggregates (A) consist of proteoglycan sub-units, hyaluronic acid, and link proteins. Most GAGS appear in the tissues in the form of proteoglycan sub-units. (Adapted from Rosenberg.[183]).

Until recently, both tendons and ligaments were grouped together as dense, regular connective tissue.[15, 37, 132] However, histologically, there were many dissimilarities between them. In analyzing the rabbit's knee ligaments and tendons, Amiel et al[15] found consistencies on gross inspection. However, on closer examination, tendons were found to contain only Type I collagen, less DNA, less GAG content, and different collagen cross-linking patterns than were found in ligaments. Ligaments contained Type I and Type III collagen, larger fibroblasts with more DNA, a different collagen cross-linking pattern, and more GAG content than the tendons. Of the ligaments examined, the cruciate ligaments contained the highest concentration of DNA and GAG content.

The fibrous capsule and ligaments provide a passive supporting structure for the joint and assist in guiding direction of movement between the two articulating joint surfaces. The more peripheral ligamentous fibres attach to the periosteum, while the more central ligamentous fibres attach to bone. This latter attachment is a transitional insertion through fibrocartilage to mineralized fibrocartilage and finally to bone.[3, 7, 45, 77] In the rabbit, the fibrous capsule and collateral ligaments are continuous with the periosteum, while the cruciate ligaments insert primarily into the transitional zones to the bone.[3]

The synovial fluid lubricates not only the articular cartilage and meniscii, but also the ligamentous structures of the joint. Motion has been shown to enhance the flow of synovial lubrication and nutrition to each of these structures.[3]

Amiel et al[15] in analysing the chemical composition of a normal ligament by weight revealed that two-thirds of the total weight was comprised of water. Of the remaining dry weight, 70 to 80 per cent consisted of collagen, the majority of which was Type I, with a smaller percentage of Type III. Three to 5 per cent of dry weight consisted of elastin, and 0.5% was comprised of GAGS. The remainder of ligamentous substance consisted of enzymes, glycoproteins, lipoproteins, and DNA from cells.[3, 15, 77]

It is thought that the presence of elastin in ligaments (absent in tendons) compensates for the fairly rigid proximal and distal attachment points of ligaments.[3] GAGS are important not only for their viscous properties, but also for their ability to bind with water. For example, the hyaluronate molecule occupies a hydrodynamic volume 1,000 times greater than if it were in the corresponding unhydrated state. [238]

A ligament or tendon can be hierarchically organized (Figure II-2) into a "structural system"[45]:

i) A collection of collagenous fibres form a primary fibre bundle[45] (subfascicular unit[60]). Surrounding this primary fibre bundle is an endotenon[45] (endotendineum[132]) sheath consisting of loose areolar connective tissue.

ii) A group of primary fibre bundles (3 to 20 subfasciculi) together form a fascicle.[45] The sheath surrounding the fascicle (epitenon[45] or epitendineum[132]) consists of a denser connective tissue than that found within the endotenon.

iii) A group of fascicles together form a tendon or ligament. The sheath surrounding the tendon or ligament (paratenon[45] or peritendineum[132]) is thicker than the epitenon, surrounds the entire tendon or ligament, and also blends with the epitenon.

iv) The collagenous fibre is further subdivided into a parallel aggregation of fibrils.

v) Each fibril (diameter 0.3 to 0.5μm[132]) consists of microfibrils which are further subdivided into fibrillar units or macromolecules of tropocollagen.

vi) Each microfibril (diameter 65nm[132]) consists of five parallel rows of tropocollagen molecules.

vii) Each tropocollagen molecule (diameter 1.5nm[129]) consists of three polypeptide chains (alpha chains), with each α chain consisting of approximately 1,000 amino acids. *Inter*molecular crosslinks extend between tropocollagen molecules at non-helical (carboxy-terminal and amino-terminal ends) and helical (teleopeptide) regions. *Intra*molecular crosslinks extend between polypeptide chains (rich in glycine and hydroxyproline).[129] At the tropocollagen molecular level, intermolecular and

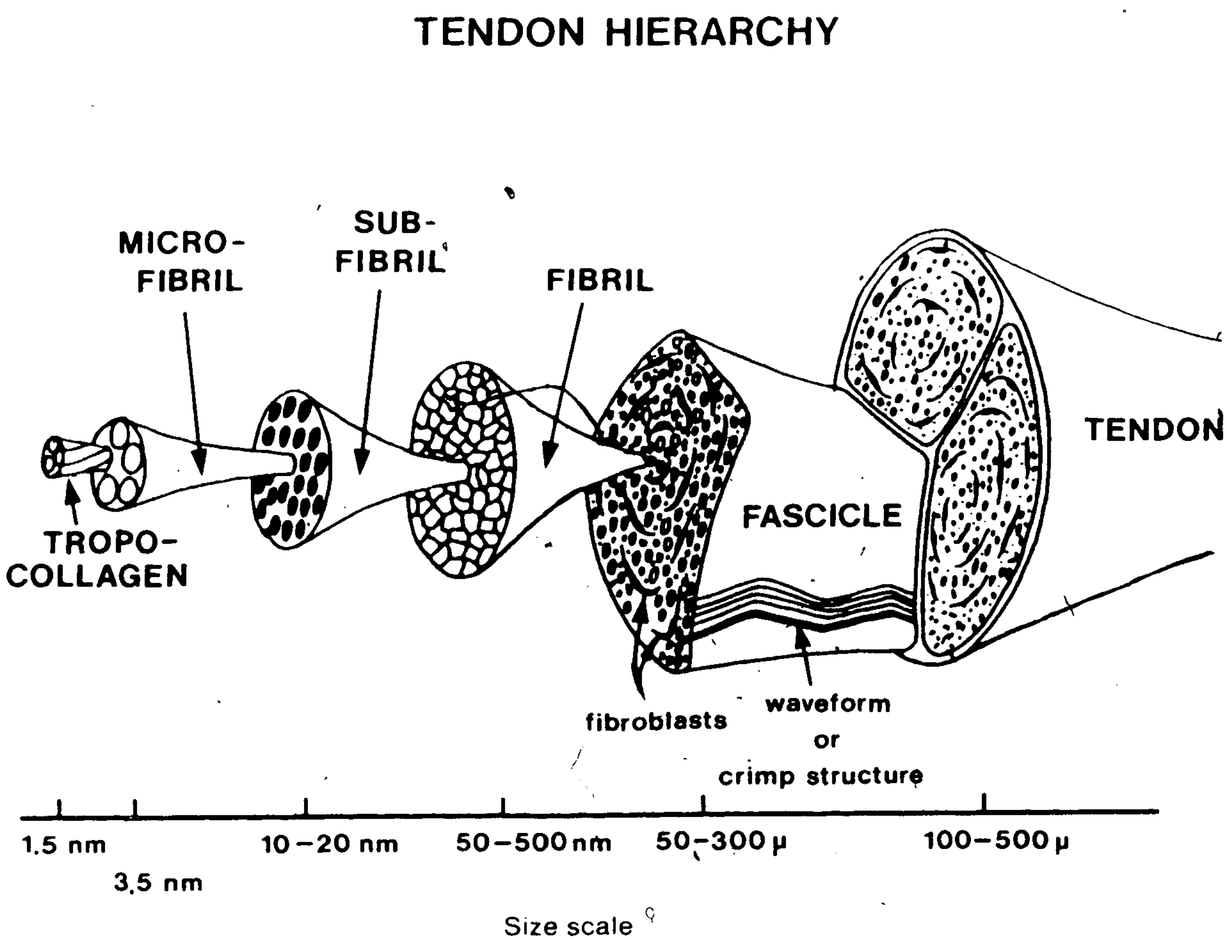


FIGURE II-2 Hierarchical Organization of Ligament or Tendon. See text for further details. (Adapted from Butler et al. 45).

intramolecular crosslinks (Figure II-3) are affected by stress-deprivation (immobilisation) in that organized deposition of these crosslinks is greatly influenced by motion.[8]

Danylchuk et al[60] noted that most collagenous fibres of the surrounding sheaths (endotenon, epitenon, paratenon) were running in a perpendicular direction to the long axis of the ligament. Furthermore, the epitenon presented in a randomly coiled manner along the length of the fascicle and "appeared to converge and bifurcate".[60]

In studies by Akeson et al, [4, 5, 9, 10, 11] the reactions of the ligamentous tissues to stress-deprivation and stress-enhancement were biochemically analyzed. Collagen content was estimated by determination of hydroxyproline level concentrations (amino acid residues within the polypeptide α chains). Chondroitin sulfate and keratan sulfate contents were either estimated or determined by analyzing the concentrations of the known sugar residues.[183] The relationship between biomechanical and biochemical changes as a result of stress-deprivation and stress-enhancement are discussed in the appropriate following sections.

2.3 Normal Biomechanical Properties of Tendons, Ligaments, and Joint Capsules

a. Introduction

The mechanical properties of ligaments, tendons, and joint capsules are usually described in terms of tensile stresses, tensile strains, and strain rate. These tissues are classified as viscoelastic: viscous depending on the rate of strain and elastic depending on the amount of strain. [45, 48]

Stress, defined as the "internal force per unit of cross-sectional area",[45, 48] is stated in equation form as $\sigma = \frac{F}{A_0}$ where F is the total force supported by the soft tissue and A_0 is the original cross-sectional area of the tissue. Tensile stresses (stretching or elongation forces) are exerted perpendicularly to the cross-sectional area of the involved tissue.

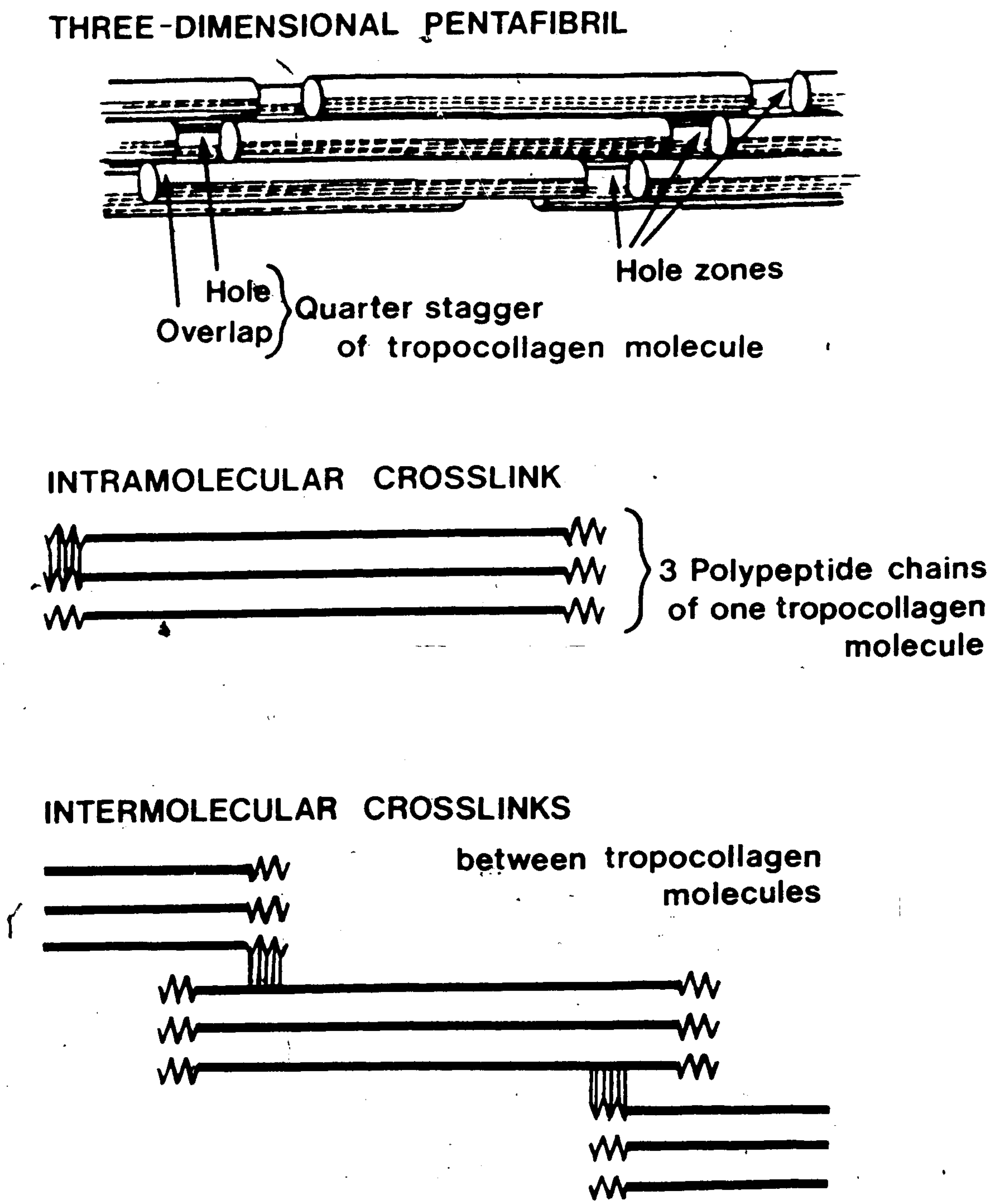


FIGURE II-3 Intermolecular and Intramolecular Crosslinks. See text for further details. (Adapted from Lane.[129]).

A. The quarter staggering relationship of tropocollagen molecules forms a microfibril.

B. At the intramolecular level, stability of the 3 polypeptide chains for each tropocollagen molecule is enhanced by intramolecular cross-links.

C. Intermolecular crosslinks extend between tropocollagen molecules.

When dealing with biological soft tissues, the equation

σ_T (True stress) $= \frac{F}{A}$, where A is the current cross-sectional area of the tissue, is more accurate

than the previously described equation for nominal stress, $\sigma = \frac{F}{A_o}$. Nominal stress is best reserved for more rigid engineering materials (Budney D: personal communication, 1987).

Small tensile strain, defined as the change in unit length of the soft tissue subjected to a load, is stated in equation form as $\varepsilon = \frac{\Delta l}{l_0}$ where Δl is the elongation length due to tensile force and l_o is the original length. The formula for small tensile strain is used whenever the amount of exerted strain is exceedingly small, as may be the case when elongation is 1 to 2 per cent. However, with a large tensile strain, such as might occur in a joint capsule or ligament taken to failure, large strain is expressed as $d\varepsilon = \frac{\Delta l}{l} = \frac{\Delta l}{l_0 (1+\varepsilon)}$ where $d\varepsilon$

is the increment in strain, l is current length, Δl is elongation length due to tensile force, l_o is original length, and d is a small change.

Furthermore, this large or true strain can be further expressed as

$$d\varepsilon_T = \frac{dl \quad \text{(small length change)}}{l \quad \text{(current length)}}$$

$$\varepsilon_T = \int_{l_0}^{l} \frac{dl}{l}$$

$$= ln \quad (1 + \varepsilon),$$

where ln is the natural log.

The equations for true or large strain can more accurately reflect changes taking place at a specific region, at any location, within the stress-strain curve. For example, with a ligament elongated

to 50 per cent beyond its resting length, the equation could analyze the area between 30 to 35 per cent elongation

Because the collagenous fibre bundles of tendons and most ligaments are parallel, these fibres can best support uniaxial tensile stresses. Whether under static or dynamic joint loading (at rest or during motion), the tendons and ligaments maintain their uniaxial load direction[3] (Figure II-4). However, the irregularly arranged collagenous fibres of the capsular ligaments are densely packed into bundles either interweaving or alternatingly angled with neighbouring bundles.[3, 132] Akeson et al[3] likened the criss-crossing 'weave' of these collagenous bundles to that of a 'nylon hose pattern'. Under tensile loading, the fibres were recruited plus elongated in the direction of the load. The resulting deformation was made possible only by the movement permitted at the 'fibre-fibre intercept points' of collagenous bundles[3] (Figure II-4). In addition, it has been suggested that the connective tissue sheaths (endotenon, epitenon, paratenon) must have a binding function rather than a tensile function, because their fibre direction is at right angles to the long axis of the tendon or ligament.[60]

b. Force-elongation Behaviour (Load-elongation Behaviour) of Tendons, Ligaments, and Joint Capsules

During tensile testing, ligaments, tendons, or sections of joint capsule are elongated or extended to failure, at a set rate, while a continuous record is made of the changes in force.

i) Force-elongation curve (load vs elongation time)

Noyes et al[160, 166] tested the anterior cruciate ligament in the rhesus monkey on an Instron testing device. All the structures of the knee were carefully removed, leaving only an intact cruciate ligament attached to the distal segment of the femur and to the proximal segment of the tibia. To permit a fairly "uniform loading" of the entire anterior cruciate ligament, the angle between tibia and femur was analogous to 45 degrees of knee flexion. With a constant strain rate, the change in time was proportional to the change in elongation.

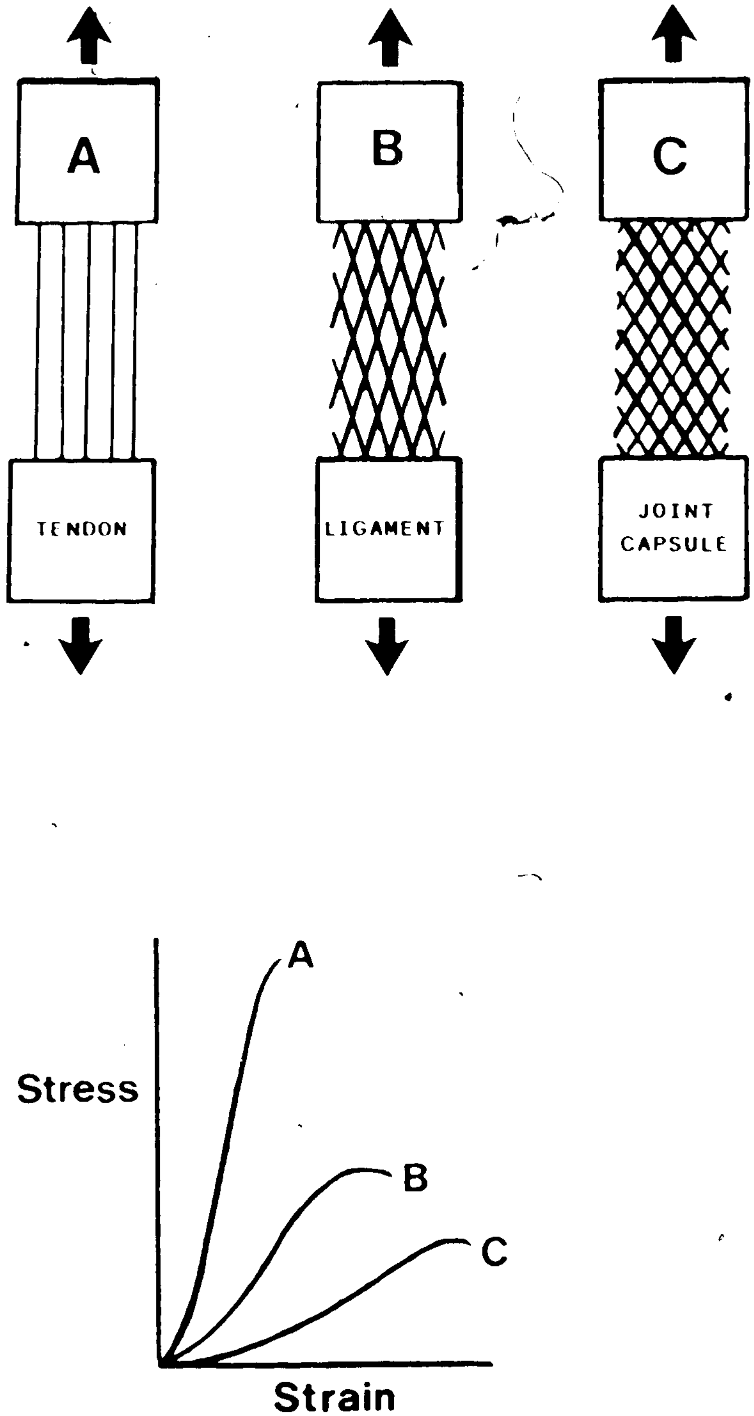


FIGURE II-4 Tensile Loading for Tendon (A), Ligament (B), and Joint Capsule (C). The upper diagram illustrates the predominant fibre direction present under tensile loading while the lower diagram illustrates the resultant stress-strain curve. (Adapted from Viidik.[237]).

On the resultant force elongation curve, the "initial concave toe region" (Region I) was followed by a fairly linear region (Region II) up to the "first significant failure" or point of linear load (Figure I-4). At this point, it was estimated that the ligamentous preparation was just entering the "major failure region". In Region II, there could be one significant failure only or a series of sequential failures. In Region III, subsequent failures, as indicated by decreases in the load, occurred in an unpredictable manner until the load dropped to zero.[160, 166] Once the point of maximum load within the major failure region was attained (Region IV), complete failure or loss of "load-bearing ability" of the ligaments occurred rapidly.

To summarize, the factors measured from the resultant force-elongation curve included stiffness or slope in the linear region, linear load (at the end of the linear region), maximum load at failure, strain to maximum load, strain to failure at maximum load, and energy absorbed at failure (the entire area beneath the curve) (Figure I-4).

The anterior cruciate-ligament failed after an elongation of approximately 57 per cent beyond its resting length. The loss of continuity of the ligament was observed from 80 to 100 per cent elongation.[160, 166] At a fast strain rate, the bone-ligament-bone preparation not only failed at a higher maximum load than the slow rate specimens, but also at an increased strain, with the absorption of more energy.[45]

According to Butler et al,[45] the initial 'toe region' (Region I) was thought to relate to a change in structure of the collagenous fibril from a crimped to a more straightened, parallel pattern (Figure II-5). With continued elongation, a 'stiffer' tissue was produced and thus an increased force was required to maintain a similar elongation. At the end of Region I, the strain or percentage elongation of the tissue, ranged from 1.5 to 4 per cent. With unloading of the force in Region I, an elastic response occurred in that the wavy pattern of the collagenous fibres was restored, as was the original length of the tissue.[45]

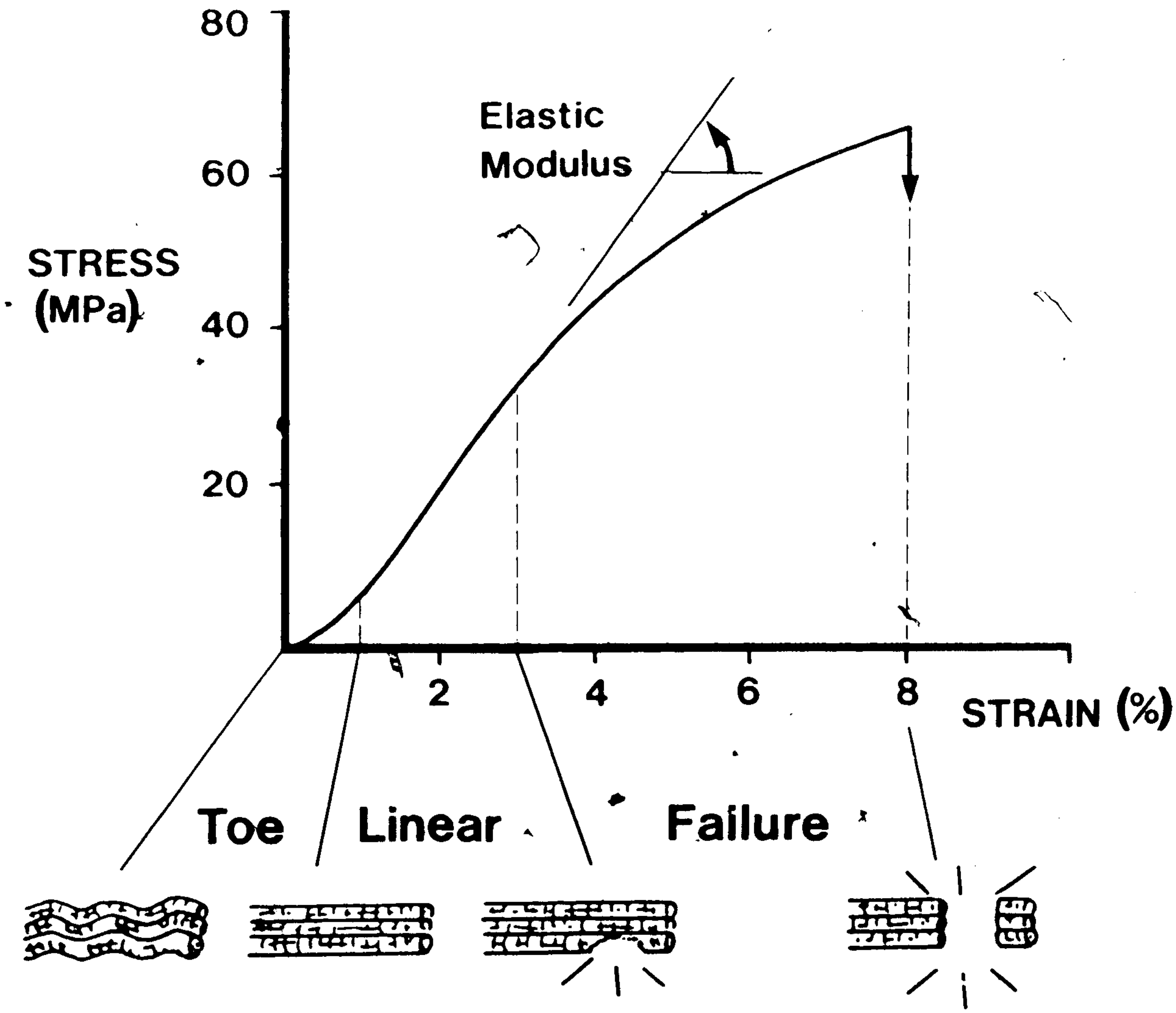


FIGURE II-5 Stress-strain Curve for Collagen. During tensile loading of collagenous fibres within Region I the fibres, which were previously wavy, straighten. At the end of the linear Region II, the first sign of failure occurs with complete failure occurring at the end of Region IV. Elastic modulus (slope in the linear region) is a measure of stiffness. (Adapted from Butler et al.[46]).

Furthermore, linear Region II illustrated the effect of further elongation of the collagenous fibres which, tested in isolation, exhibited strain rates varying from 2 to 5 per cent. However, when entire ligaments or tendons were tested, strain levels as high as 20 - 40 per cent could be attained. Before the end of Region II, small reductions in force could often be observed, most likely due to the early failure of a few over-extended fibre bundles (sequential failures). The point of linear load was significant because it indicated that the first major failure of fibre bundles had occurred.[45]

Following further elongation of the ligament or tendon, the curve could level off shortly after the point of linear load, fail rapidly after attaining maximum load, or fail in consecutive steps after attaining maximum load. Because ligaments and tendons insert on areas, not on points, it is expected that serial failures would occur as the load shifted from one set of overstretched fibres to another set, until they too failed.[234]

Kennedy et al[120] studied human medial collateral and cruciate ligaments which were excised in their entirety within twelve hours post-mortem. At point of maximum load, the ligaments appeared intact by macroscopic examination despite elongation of the ligament by 20 to 30 per cent beyond its resting length. However, using the scanning electron microscope, there was evidence throughout the ligament of disruption and disorganization of the collagenous fibrils, especially in the deeper fibrils. Applying load beyond ultimate failure caused complete disruption of the fibres. The microscopic appearance of fibrils thus disrupted, appeared similar to that of fibrils tested just to the point of ultimate failure; however, disorganization of the fibrils was much greater.[120]

According to Butler et al,[45] the shape of the load-elongation curve (whether testing ligament, tendon, or joint capsule) varied, depending on the original length of the testing tissue, its original cross-sectional area, the orientation of fibres in relation to the joint, and the "tissue microstructure".[45] Ligamentous tissues with similar cross-sectional areas but different lengths displayed similar maximum loads. However, longer specimens underwent a greater deformation because the tissues stretched further before failure. These tissues also exhibited less stiffness, as shown by a decrease in slope of the curve in the load-elongation curve. Similarly, ligamentous tissues with a larger cross-sectional area had an increased number of collagenous fibres, and were thus able to withstand a larger maximum load than ligaments with a smaller cross-sectional area.[45]

Grood et al (as quoted by Butler et al[45]) studied the load-elongation behaviour patterns for bone-ligament-bone specimens of the rhesus monkey's medial collateral ligament and medial capsule of the knee (anterior and middle portions). The medial collateral ligament (as compared to the capsule) reached a much higher maximum force, followed by a markedly more abrupt failure (Figure II-6). At the point of complete failure of the collateral ligament, the entire capsule was supporting only half of the collateral ligament force. 'Serial' or 'sequential' failure was most marked in capsular preparations (Figure II-6).

Akeson et al [3] felt that orientation of the capsular fibres in the direction of tensile load was only possible when movement was permitted at the 'fibre-fibre intercept points' of the 'criss-crossed' nylon hose pattern. This lengthening of fibres along the line of stress created the initial toe region. This toe region for the joint capsule as a whole was longer than that of the remaining ligaments, especially the more "loosely woven" posterior portion of the joint capsule [3] (Figure II-4).

ii) Stress-strain curve

A stress-strain curve[45] is created by adjusting the load-deformation curve by dividing the applied force by original cross-sectional area (yielding tensile stress), and then dividing the deformation by initial length (yielding tensile strain) (Figure II-5). The mechanical parameters calculated from this stress-strain curve consist of elastic-modulus (slope in linear Region II), maximum stress at failure, strain to maximum stress, and energy density (area beneath the curve). Stress-strain curves, however, can only be calculated if accurate measurements can be made of the cross-sectional area and length of the tissues (ligamentous or tendinous) in both original and final dimensions.

The results from the stress-strain curve (Figure II-5) are similar to (with regards both to the shape of the resultant curve and the resultant obtained information [45]), but have greater accuracy than

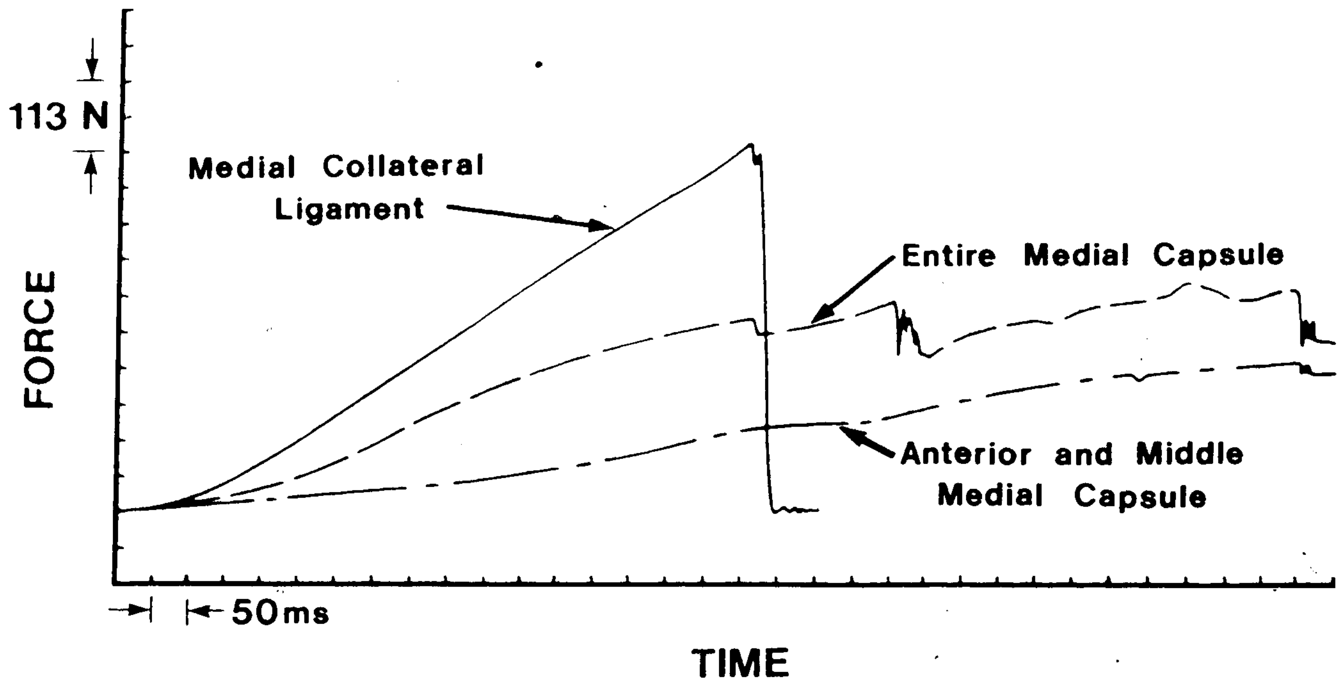


FIGURE II-6 Force-time (Load-elongation) Curve for the Medial Collateral Ligament and Medial Joint Capsule of the Rhesus Monkey. The medial collateral ligament fails abruptly at a much higher maximum force than the joint capsule. Sequential or serial failure is evident for both capsular preparations. (Adapted from Butler et al.[45]).

those from the load-elongation curve (Figure I-4). If the original and final dimensions both for the cross-sectional area and length of tissues are known, then the stress-strain curve can more accurately reflect the changes taking place under tensile loading than the load-deformation curve alone.

Woo et al [252] have noted however that the strain of the mid-ligamentous substance is consistently much less than the deformation of the bone-ligament-bone specimen. It was found that the deformation at or near to the ligamentous insertion sites was greater than the deformation in the mid-substance of the ligament due to the fact that the ligamentous insertion points on the bone were variable and covered a larger area. The rabbit specimens in particular showed a "large variation in regional strains at failure as the averaged tibial strain was approximately twice that of the femoral strain" [252] with the result of more tibial avulsion fractures on testing.

iii) Hysteresis

Butler et al [45] has described the formation of a hysteresis loop for a bone-ligament (tendon)-bone specimen under tensile loading. The specimen was stretched to a predetermined peak strain and was allowed to return to its original starting length at a constant displacement rate. This test commenced either with the tissue at its original length or with some loading already present. The area between the resulting loading and unloading curves indicated the amount of energy lost during this hysteresis test. With "repeated cycling" of the tissue "to the same peak strain",[45] the area of the loop could be reduced (Figure II-7). Furthermore, with repeated cycling, the curves successively shifted to the right (Figure II-7).[237] Both Butler [45] and Viidik [234] interpreted the forming of a hysteresis loop as an indication of the viscous properties of tendons and ligaments.

c. Torque-angular Displacement Curve

Woo et al [254] passively cycled the rabbit's control knee joint through a predetermined range of motion of flexion-extension-flexion within physiological limits of the normal range on an arthrograph. This passive cycling to a specific range 50°-80° and back to the starting point 80°-50° on the

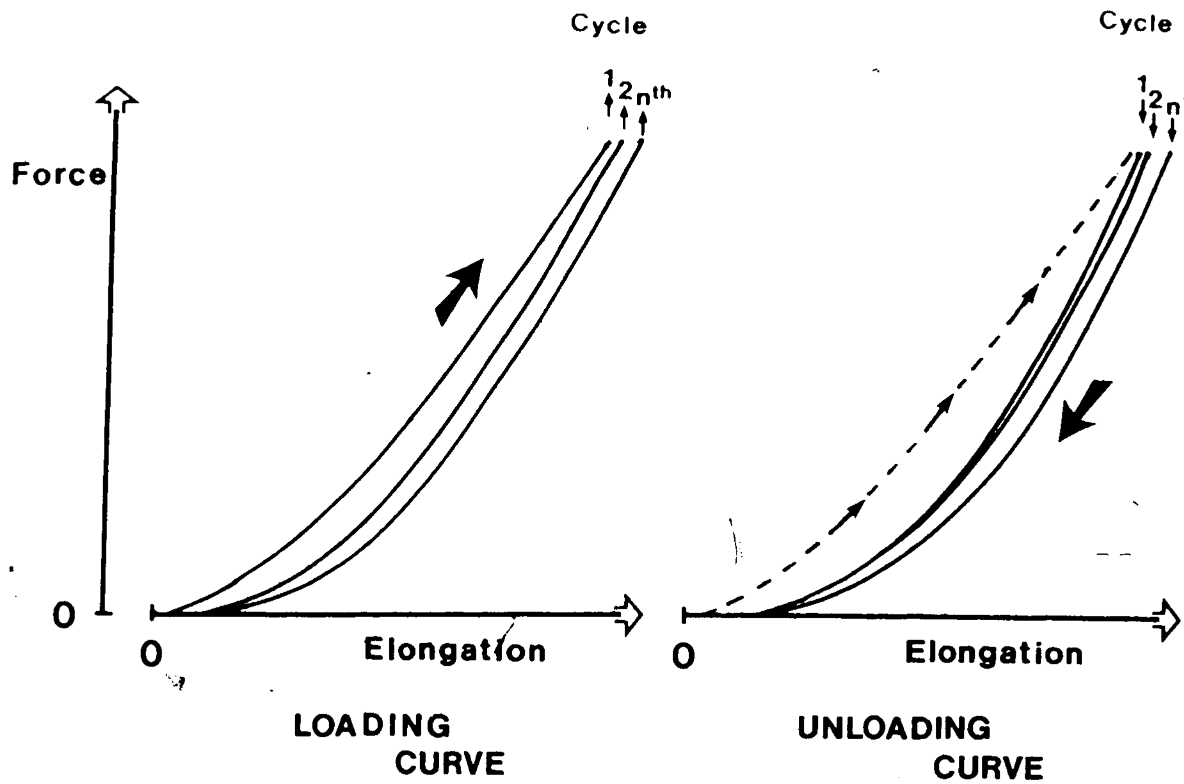


FIGURE II-7 Repeated Hysteresis Loops on a Force-elongation Curve for a Bone-Ligament-Bone Specimen. During tensile loading (loading curve), the specimen is elongated within the linear region to a predetermined peak strain at a constant rate and then returned to its original starting length (unloading curve). These cycles are repeated from one to two to **n** times. The area of these repeating loading-unloading cycles decreases over time. As well the curves successively shift to the right. (Adapted and modified from Viidik.[237]).

arthrograph was documented by an X-Y recorder to yield a torque-angular displacement curve (Figure II-8). Prior to the actual testing, the rabbits were sacrificed and the lower limbs disarticulated with muscle attachments divided above and below the knee joint. In these preparations, resistance to movement was due primarily to the ligaments, joint capsule, and articulating joint surfaces.

The slope of the curve was proportional to the stiffness of the joint and the enclosed area of the hysteresis loop represented the energy required for one complete cycle of motion. For a control (normal) joint the stiffness was minimal as depicted by the absence of slope and the energy lost was minimal as depicted by the minimal enclosed area (Figure II-8).

2.4 Effect of Joint-Immobilisation on the Biomechanical Properties of Tendons, Ligaments, and Joint Capsules

a. Joint-Immobilisation

During immobilisation of a joint, changes occur in both the periarticular soft tissues surrounding the joint and in the articular cartilage with its underlying subchondral bone. There is a consistent fibrous, fatty proliferation of the connective tissue within the joint space.[6] These changes not only depend upon time,[6, 10, 74, 130] but also appear to depend upon the angle of the involved joint during immobilisation.[74, 130, 233]

As a readily available subject, the rabbit has been utilized by various researchers [6, 10, 17, 74, 130, 150, 229, 233, 254] for studies on the effects of joint-immobilisation. The fully flexed position of the rabbit's knee has been reported at 170° with full extension at 0 degrees.[150, 229] Salter reported the resting position to be either 130[186] or 140[188] degrees of flexion. Human joints are immobilised throughout the range 0° - 90° depending on the pathology and treatment.

Finsterbush and Friedman[74] immobilised rabbits' knees in a position of 15 to 20 degrees flexion. The trunk and hind limb were positioned in a Plaster of Paris spica for a time period ranging

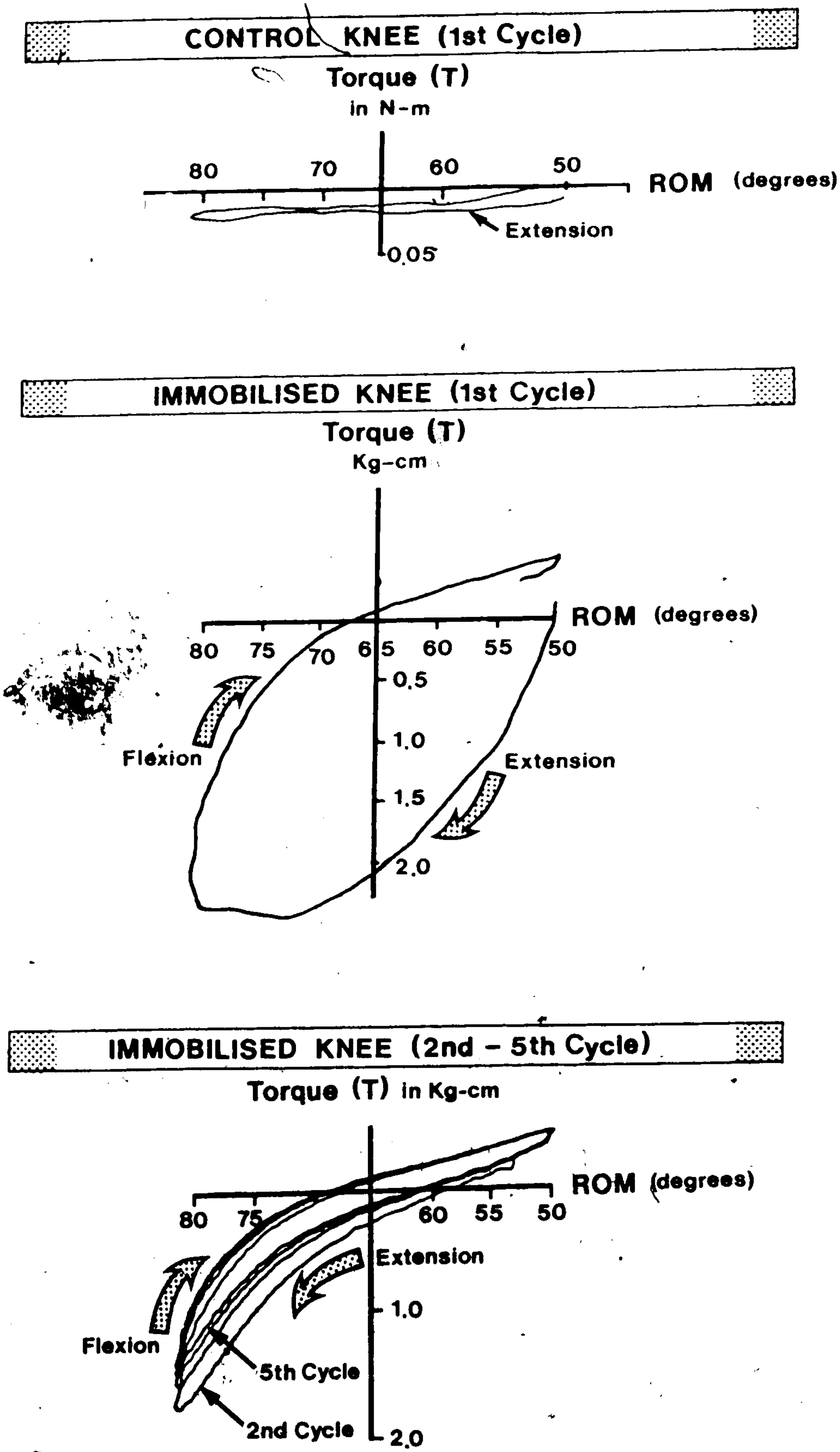


FIGURE II-8 Torque-angular Displacement Curves for Control and Immobilised (Contracture) Rabbits' Knees. See text for further details. (Adapted and modified from Woo et al.[254]).

from two to eight weeks. The second to eighth week were marked by a gradual increase in capsular thickening and a marked restriction of joint movement.

Langenskiöld et al[130] immobilised rabbits' knees in the fully extended position for varying time periods. It was noted that flexion up to 20° was possible within the splint. Following six days of immobilisation, histological sections were taken and revealed thickening of the collateral ligaments, the infrapatellar tendon, and the joint capsule. After 14 days of immobilisation, osteophyte formation was noted on the tibial joint surfaces. Both Videman[229] and Langenskiöld et al[130] found that knees immobilised in full flexion, for time periods from four to ten weeks, showed little evidence radiographically or macroscopically of joint degeneration.

The development of a joint contracture, involving a tightening or shortening of skin, fascia, muscle, and joint capsule, results in a significant loss of ROM. The normal elasticity of the immobilised tissues is affected by abnormal adherence of collagenous fibres to surrounding tissues. Consequently, profound biomechanical and biochemical changes occur in the periarticular connective tissue.

b. Biomechanical Changes

i) Torque-angular displacement curve

Woo et al[254] examined biomechanical changes occurring in rabbit knee joint contractures. The experimental procedure for immobilisation of the rabbit's knee joint was repeated by the same group of workers from 1973 to 1985.[6, 10, 14, 16, 17, 254] In each case, the knee was maintained for nine weeks in a position of full flexion by means of a surgically-inserted Steinmann pin. Following this immobilisation, the rabbits were sacrificed and the lower limbs disarticulated with muscle attachments divided above and below the knee joint. In these preparations, resistance to movement was due primarily to the ligaments and joint capsule.

The immobilised knee was tested on the arthrograph in flexion-extension-flexion, which was documented on the X-Y recorder to yield a torque-angular displacement curve. The slope of this curve was assumed to be proportional to the stiffness of the joint, while the area enclosed by the hysteresis loop represented the energy needed for one cycle of movement (Figure II-8).[254]

Both contracture and control knees were flexed and extended through a small range (50° - 80°) for a total of five cycles and through a larger range (45° - 95°) for another five cycles. In this instance, 180 degrees was regarded as the fully extended position. The amount of torque required to extend the contracture knees — from either 50 to 80 degrees or 45 to 95 degrees — during the 1st cycle was four times greater than that required for the control knees. During the 5th cycle, the torque for 50 - 80 degrees decreased to two and a half times greater for contracture knees, while for the 45 - 95 degree range the torque decreased to three times.[254] The area of hysteresis for the 1st cycle of flexion and extension (50° to 80° to 50°) was 10 times greater for contracture knees as compared to the control knees. For the cycle 45 to 95 degrees, the area of hysteresis was six times greater.[254]

An earlier study by Akeson et al,[10] showed that the strength of the contracture increased in a linear fashion in relation to the studied immobilisation period (one to nine weeks) (Figure II-9). Following one week of immobilisation, the amount of torque during the 1st cycle (50° to 80°) was three times greater for contracture knees as compared to the control knees, while in the 45 to 95 degree range (1st cycle), the torque decreased to two times. The area of hysteresis for the 1st cycle (50° to 80°) was four times greater for the contracture knees, while for the 45 to 95 degree range (1st cycle) the area of hysteresis decreased only two and a half times.

ii) Force-elongation curve

Noyes[158] and Noyes et al[166] studied the immobilisation effects of a total body plaster cast on the rhesus monkey's anterior cruciate ligament. Following eight weeks of immobilisation, these bone-ligament-bone specimens were tested to failure at a fast rate of deformation, as previously described.[160] As depicted on the force-elongation curve (Figure II-10A), maximum load at failure was decreased

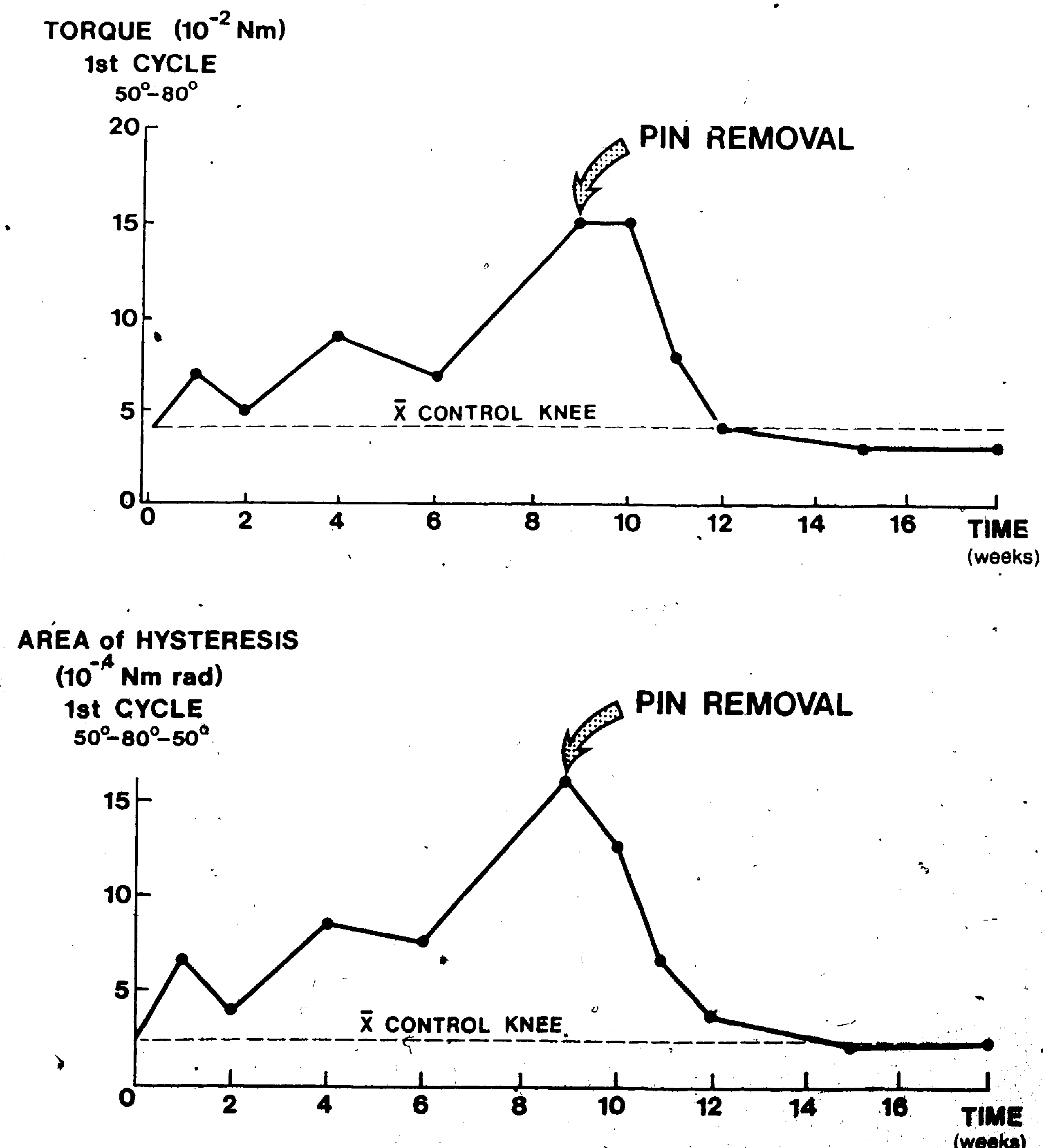


FIGURE II-9 Time Sequence for Torque and Area of Hysteresis in the First Cycle. The results shown are from hysteresis loops of immobilised rabbits' knees in weeks one through nine, and remobilised rabbits' knees in weeks ten through eighteen). See text for more details. (Adapted from Akeson et al.[11]).

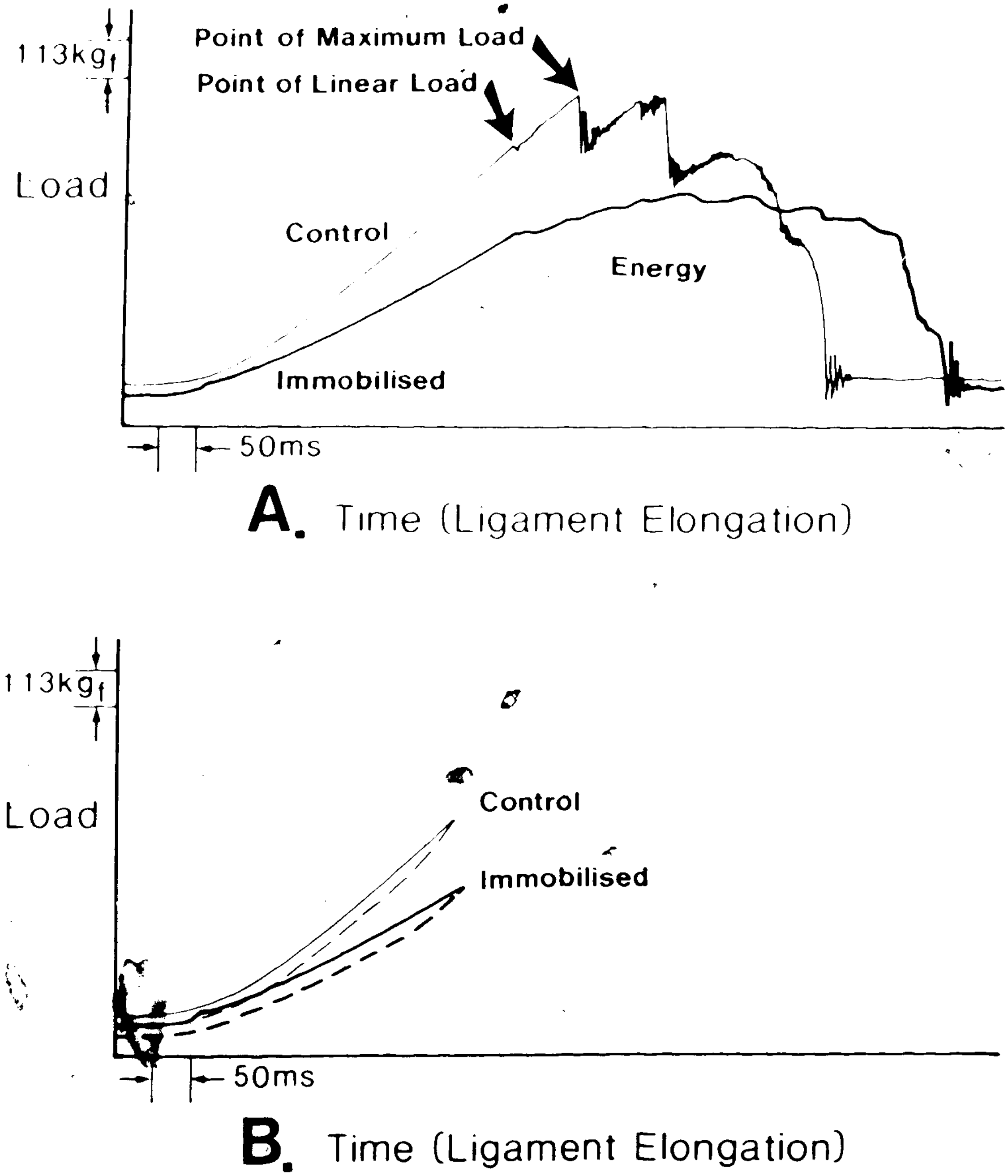


FIGURE II-10 Load-elongation Curves (A) and Hysteresis Loops (B) for Control and Immobilised Tibia-Anterior Cruciate-Femur Specimens of the Rhesus Monkey. See text for more details. (Adapted and modified from Noyes et al.[166]).

TABLE II 1

Summary-Comparisons of Immobilisation and Remobilisation Groups of Rhesus Monkeys from Studies by Noyes [160, 162, 166]

Subject Group	Stiffness*	Maximum Load at Failure*	Energy Absorbed at Failure*
Control (normal)	100%	100%	100%
Total Body Immobilisation (8 weeks)	69%	61%	68%
One Limb Immobilisation (8 weeks)	88%	88%	79%
Remobilisation (5 months)	93%	79%	78%
Remobilisation (12 months)	98%	91%	92%

* indicates percentage of control

by 39 per cent as compared to the control group (Table II-1). In addition, the energy absorbed to failure (area under the curve) was decreased by 32 per cent. The immobilised specimens showed a definite decrease in stiffness (slope) of 31 per cent.

c. Correlation Between Biomechanical and Biochemical Changes as a Result of Joint-Immobilisation

In the study by Woo et al. [254] loss of GAGS in the immobilised periarticular connective tissue correlated significantly with the areas of the hysteresis loop and the torque required to extend the experimental rabbit's knee, as depicted in the torque angular displacement curve. This correlation was shown for both the small and large ranges of motion — especially during the first cycle. [254] Decreases in chondroitin-4 sulfate and chondroitin-6 sulfate (30%) and in hyaluronic acid (40%) were significant reductions in the GAG concentration of stress-deprived (immobilised) periarticular connective tissue.[6, 87] Furthermore, water content was decreased by four to six per cent as a result of the nine week immobilisation period. A decreased concentration of GAGS (especially hyaluronic acid) and water altered not only pliability, but also the elasticity-plasticity of connective tissue. With this reduction of GAGS and water, the distance between collagenous fibres became reduced.[6, 87]

Noyes et al [160] suggested that GAGS might provide a form of lubrication between individual collagenous fibrils and fibres. Viidik et al[238] suggested that GAGS could be a causative factor in the waviness of collagenous fibre bundles. Furthermore, Viidik et al [238] felt that the sheath of GAGS surrounding the fibrils possessed an abundant "mechanical extensibility" because this sheath remained intact even after the fibril was extended to failure.[238]

Akeson et al [10] felt that the synthesis of proteoglycans and collagen was modulated by joint motion.[10] Without motion, a haphazard network of anomalous crosslinks most likely developed within the immobilised periarticular connective tissue. In addition, the distance between collagenous fibres probably became reduced as concentrations of both water and hyaluronic acid were diluted. Abnormal crosslinks could act as bridges between formerly independent fibrils. Furthermore, these abnormal crosslinks could act as mechanical constraints when the immobilised joint was mobilised. In contrast, motion should lubricate and maintain the critical distance between existing collagenous fibres. As a result of motion the new collagenous fibres could be laid down along the normal lines of stress and, as a result, anomalous cross-links could be minimized.

On the arthrograph, the strongest resistance to knee extension was always observed during the 1st cycle (Figure II-8) of the torque-angular displacement curve. This large increase in joint stiffness most likely resulted both from gross adhesions between articulating joint surfaces and from anomalous intermolecular crosslinking between collagenous fibres and fibrils (in the periarticular connective tissue).[3, 254] In comparison, the load-elongation curve depicted tensile testing of previously immobilised tendons plus ligaments, and revealed reduced stiffness and a decrease in maximum load at failure (Figure II-10). However, what one is seeing is a relationship between changes of a joint complex (with effect of muscles removed) in contrast to those of a bone-ligament-bone specimen. The joint capsule with its multi-axial meshwork of collagenous fibres will behave differently than a ligament with a parallel fibre arrangement (Figure II-4).

In both situations (joint complex; bone-ligament-bone), Akeson et al [5] noted as a result of joint-immobilisation a statistically significant decrease in GAG concentration and water content as well as an increase in collagen synthesis and rate of collagen degradation. Along with the increase in collagen synthesis, a significant increase (about 30%) in intermolecular crosslinks occurred.[5] It has been suggested by Akeson et al [5] that this loss of GAGS and H_2O affected the lubricating effectiveness of these substances and resulted in a decreased flexibility at the fibre-fibre interface.[5] Amiel et al [15] confirmed that these lubrication effects and the maintenance of fibre-fibre distance were largely dependent upon levels of hyaluronic acid.[16] As part of their study, rabbits underwent a nine week immobilisation of the left knee by receiving once-weekly injections of hyaluronic acid. This treatment resulted in a considerable decrease (50%) in total 1st cycle stiffness on the arthrograph.

The cruciate ligaments appear particularly liable to biochemical changes, thought possibly to be ligament-specific.[8] Within the human anterior cruciate ligament, in particular, an abundance of loose connective tissue comprises the ligamentous sheath.[60] The ratio of area of connective tissue sheaths to the area of collagen fasciculi varies both along the length of the ligament and between individual ligaments.[60] The effect of immobilisation on these sheaths does not appear to have been mentioned in the literature.

Tensile loading of the bone-ligament-bone specimen will most likely result in a tearing of the immature cross links. With increased collagen synthesis, new collagen will be relatively immature and of a lower strength. During arthrographic testing, not only is the multi-axial joint capsule tested, but also the articular cartilage, as well as the ability of collagenous tissue to glide in relation to neighbouring collagenous tissues.

Viidik[237] stressed the importance of relating tissue geometry to its biomechanical behaviour. He thought parallel-fibred tissues (ligamentous or tendinous) would respond well to tensile loading stresses with a steep linear curve during the loading and unloading hysteresis cycle (Figure II-4A). An open meshwork with a 'main fibre direction' would respond similarly (Figure II-4B). However, the decrease in linear stiffness would be due to the geometrical configuration of the meshwork in addition to the fact that some of the meshwork fibres were cut in specimen preparation. Viidik further reasoned that the strength of the tissues without such apparent main fibre direction, for instance the joint capsule (Figure II-4C), would be dependent on the "friction, cohesion or binding" between fibres of the meshwork.[237] For additional strength, the ligamentous fibre bundles would be arranged in meshed layers.[79] He concluded that the increase in elongation capabilities and decrease in stiffness (Figure II-4C) would result from a continuing rearrangement of layers moving in relation to one another in the direction of applied stress.

2.5 Effect of Joint-Immobilisation Followed by Exercise on the Biomechanical Properties of Tendons, Ligaments, and Joint Capsules

a. Force-elongation Curve

Noyes[58] and Noyes et al[166] studied the effect of exercise following a period of joint-immobilisation. After eight weeks of total inactivity in a body cast, rhesus monkeys were divided into groups having either 5 or 12 months reconditioning (active exercise in both). At the end of the remobilisation period, femur-anterior cruciate-tibia specimens were tested to failure.

Specimens tested immediately following immobilisation showed maximum load at failure decreased by 39 per cent as compared to control specimens on the load-deformation curve. Maximum load to failure for the animals having 5 months reconditioning was decreased by 21 per cent as compared to control groups. With 12 months reconditioning, the maximum load to failure was within 9 per cent of the control group (Table II-1).

For the animals with 5 months of reconditioning, energy absorbed to failure was reduced by 22 per cent as compared to the controls. For animals with 12 months of reconditioning, the energy absorption to failure was 92 per cent, indicating a nearly complete recovery.[158] The amount of stiffness had significantly increased by 5 months (93%) with a much slower rate of increase in the last 7 months for the 12 month reconditioned group (98%)(Table II-1).

In a variation from total immobilisation regimes, Noyes et al [166] immobilised rhesus monkeys for eight weeks in a total body cast, except ~~for one~~ free lower extremity. During exercise of this free limb (exercise being food-rewarded), isometric contractions of the immobilised thigh muscles were observed. The resultant force-elongation curve for the immobilised knee was midway between the immobilised and 5 month reconditioned group as per Noyes [158] (Table II-1).

Figure II-11 illustrates the load-deformation curve for the various treatment groups of bone-anterior cruciate-bone specimens (adapted from Noyes [158]). The results for the groups — 5 months

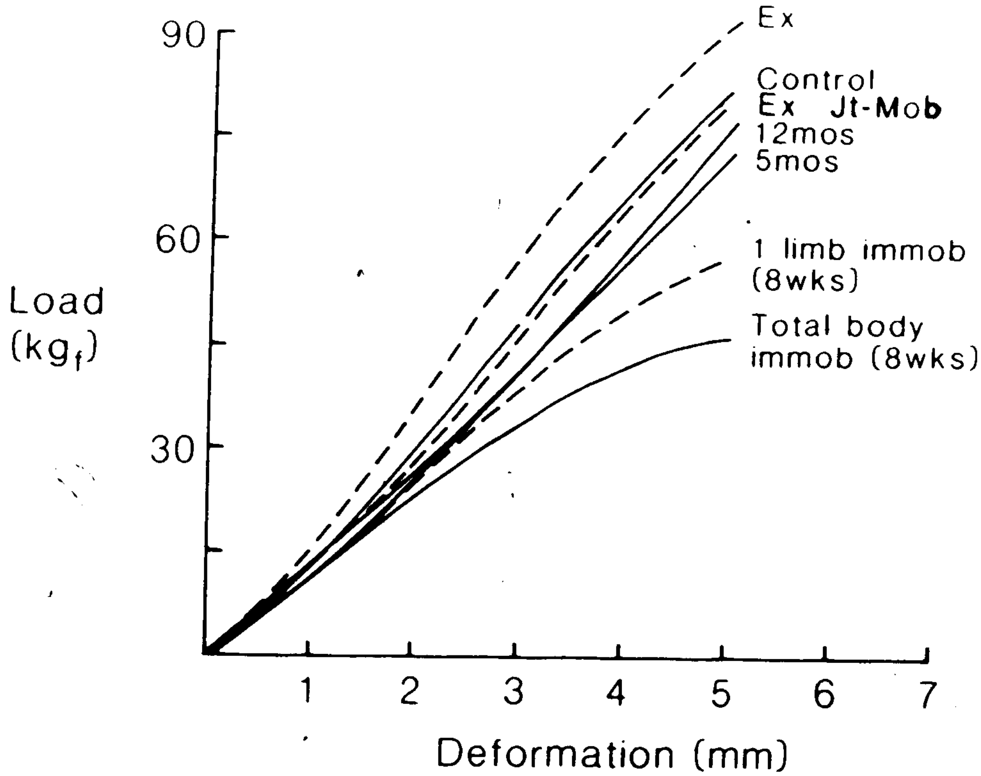


FIGURE II-11 Load-deformation Curve for Immobilised and Remobilised Bone-Anterior Cruciate Ligament-Bone Specimens of the Rhesus Monkey. Immobilised groups (total body immob and 1 limb immob) plus remobilised groups (5 months and 12 months) are detailed in the text. Both the active exercise group with joint-mobilisation (Ex Jt-Mob) and normal exercised group (Ex) are discussed in the conclusion section. (Adapted and modified from Noyes.[158]).

reconditioned, 12 months reconditioned, total body immobilised, and total body immobilised-1-free-limb — are portrayed schematically.

b. Torque-angular Displacement Curve

Figure II-9 demonstrates the gradual development of an increase in torque and area of hysteresis for an immobilised rabbit's knee from week one to week nine, and recovery from the contracture during nine weeks of remobilisation.[10, 11] Remobilisation consisted of unrestricted cage activity.

The arthrograph was used to test the remobilised rabbits' knees in the same manner as described for the immobilised knees.[254] Contracture development, with increasing area of hysteresis and increasing torque, occurred at a slower rate than did the recovery from contracture with its decreasing area of hysteresis and decreasing torque (Figure II-9). By the sixth week of recovery, both areas of hysteresis and torque measurements for the range 45 - 95 degrees were essentially equal to those of the control knees.[11] Whereas, Akeson et al[11] noted that recovery during the first two months was relatively rapid,[11] this recovery slowed to a much more modest rate and may, indeed, according to Noyes et al [158] have extended to longer than one year. [158]

c. Effect of Exercise (Without Prior Period of Joint-Immobilisation)

Woo et al [251, 253] exercised miniature swine on a motorized treadmill. Following a 12 month training period, testing of swine digital flexor tendons showed on the load-deformation curve that the ultimate load at failure was 19 per cent higher for the exercised flexors than for the controls. Similar findings were noted by Tipton [217, 220] and also by Cabaud [49] with rats.

2.6 Physiotherapeutic Joint-Mobilisation

Two physiotherapeutic approaches to treatment of joint hypomobility apply to this study: active exercises, and exercises in conjunction with joint-mobilisation of the affected joint. Specific joint-mobilisation involves either a joint-gliding (in the direction of the restricted gliding) or a traction manoeuvre, in order to restore normal joint range.[117] Joint-mobilisation is a passive, low-velocity movement of varying amplitude performed within the existing painfree and muscle spasm-free joint range.[117, 141] A Grade III joint play movement (traction or gliding) is a large amplitude movement performed from the mid-point of the range to the end of the existing joint range. A Grade IV movement is a small amplitude movement performed at the end of the existing joint range (Figure I-2).

2.7 Joint Range of Motion

The amount of motion or movement available at a specific joint is conventionally termed range of motion (ROM). The total possible range of either active or passive movement is determined by the geometry of the moving joint surfaces, as well as by limitations imposed by the joint capsule, ligaments, and surrounding musculotendinous structures.

All moving bones articulate at joints. The movement of a limb through space can be subdivided into arthrokinematics, the study of movements within the joint, and osteokinematics, the study of gross movements of bones.[140] Osteokinematic movements of active or passive knee extension, with the femur relatively stationary, involve the movement of the tibia in an anterior direction (Figure I-3). The arthrokinematic movements occurring during knee extension consist of an anterior gliding of the concave proximal joint surface of the tibia in relation to the relatively stationary convex surface of the femur (Figure I-2).

During an osteokinematic movement, a limb may be actively moved through space, for instance extension of the leg at the knee. This same movement of knee extension may be repeated passively. At the end of a passive movement, a resistance to further motion can be felt. This end-feel sensation at the extreme of a normal passive ROM can be described as capsular for knee extension, bony block for elbow extension, and a soft tissue approximation for knee flexion.[59] There are many abnormal (pathological) end-feels as well, but for the purposes of this study the only abnormal end-feel considered is that of a capsular end-feel. An abnormal capsular end-feel (at the end of the available passive range) occurs earlier in the ROM than would normally occur if a full ROM were present. The normal and pathological end-feels are similar in sensation with the abnormal end-feel offering a greater resistance due to a thickening of the joint ~~capsule~~. Grading of osteokinematic movements is determined relative to the pathological amplitude of bone movement as determined by the capsular end-feel (Figure I-3).

Passive joint play or accessory joint movements are performed by the physiotherapist to further examine the quality of arthrokinematic movements. These accessory movements of traction and joint-gliding form the basis of joint-mobilisation techniques. Grading of arthrokinematic movements is determined relative to the pathological amplitude of available traction or gliding at the joint as denoted by the capsular end-feel at the end of the available range of passive motion. It has been suggested that traction or separation of the two articular joint surfaces produces a stretching of the joint capsule.[40, 54, 55, 172] Gliding joint-mobilisation involves a gliding in the direction of the limitation of movement. For instance, to help restore a loss in knee extension, the tibia is passively moved in an anterior direction in relation to the stationary femur. It has been suggested that this gliding motion specifically restores the lost joint gliding, lubricates the joint surfaces, and stretches both the joint capsule and periarticular joint structures.[40, 124, 172, 173, 267]

2.8 Measurements of Joint Stiffness

To provide consistency in understanding joint stiffness, the following definition by Thompson et al [215] will be used throughout the text. Joint stiffness is defined as "the resistance to passive motion at a joint throughout the normal range of motion in the usual functional plane". Physiotherapists measure joint stiffness both subjectively and objectively. However an accurate and reproducible quantification of the amount of joint stiffness remains inadequate at the present time.

a. **Subjective Measurements**

Rhind et al[181] attempted to investigate definitions of joint stiffness as characterized by patients. According to this study, the words commonly used by patients to describe joint stiffness could be classified under three different, major descriptors: difficulty of movement, pain, and sensations. From this sample of 100 patients, the words chosen for these three descriptors were i) 'difficulty of movement': limited movement, rigid, stuck, inflexible, immobile, solid, fixed; ii) 'pain': painful, aches, hurts, sore; and iii) 'sensations': tight and tense. Most commonly chosen were 'limited movement', followed by 'painful'. This study made it evident that a patient complaining of stiffness could be referring to joint stiffness, pain, or a combination of pain and stiffness.

In addition, patients were exposed to three different types of scales rating degrees of stiffness: a visual analogue marking scale, a numerical rating scale, and a 5-point verbal scale (Appendix A). By means of these scales, patients appeared able to assess the severity of their stiffness, despite the fact that they were unable to define the symptoms of stiffness in a consistent manner.

In earlier studies by Ingpen and Kendall[102] and Scott,[193] patients were often found to be incapable of distinguishing between joint stiffness and pain. In a series of studies exploring the significance of stiffness in the arthritic hand and the accurate measurement of stiffness, direct relationships were found between grip strength and pain, grip strength and joint stiffness, and grip strength and increase in morning hand volume. Grip strength was found to be decreased in the presence of pain, joint stiffness, or increased morning hand volume.[193, 221, 256] In the diagnosis of rheumatoid arthritis, morning stiffness heads the list of eleven diagnostic criteria as set out by the American Arthritis Association.[107, 147, 193] However, if patients are incapable of accurately distinguishing between joint stiffness and pain these subjective scales for rating degrees of stiffness must be questioned for their overall validity.

b. Objective Observations of Activities of Daily Living

A Guttman scaling technique (Appendix A) is often used to arrange activities in order of difficulty. In terms of the scale, inability to perform one activity implies that it would be impossible to perform another activity further up the scale; however, no difficulty should be experienced with performing an activity further down the scale. The World Health Organization attempted to relate arthritic pain and restriction of joint movement with difficulty or inability to perform various activities of daily living (ADL).[22] However, problems with design of a general functional index have led to discussions of the need for specific functional indices, one for each particular group of patients. [22, 44]

Examples of ADL observations of varying activities used to assess the functional status of an arthritic patient are outlined by Melvin,[147] Trombly,[223] and Jette.[105] Although assessment-of-function forms have been used for years, the ROM required for specific activities is incompletely documented[23]. A Guttman scaling technique should theoretically be more accurate than previously described subjective measurements of joint stiffness. However, each developed scaling technique needs to be scientifically evaluated and scrutinized.

c. Goniometric Measurements and Mobility Testing.

The ROM at a joint is conventionally described by measuring degrees of range with one of the various types of goniometers.[42, 151, 152, 157, 222] ROM norms for all joints of the body can be found in one of the first source documents by the American Academy of Orthopaedic Surgeons (1965).[112] As a result of physiotherapy, there may be a clinically significant increase in ROM, which may or may not be accompanied by an improvement noted in activities of daily living. Throughout the literature on both the various means of documenting joint ROM and observations of activities of daily living (as correlated with ROM), no mention has been made of either joint stiffness or the measurement of such.[105, 151]

Attempts to classify the joint mobility of patient populations into hypermobility (hyperlaxity of joints)[32] or hypomobility (hypolaxity of joints) have been extensively detailed by Carter and Wilkinson [51] and, more recently, by Beighton et al.[30] These joint mobility measurements give an overall impression of the total range of motion present throughout the body and compare one individual with another individual. However, these are merely more generalized range of motion observations, because only certain joint movements are examined -- hyperextension of the fifth finger, elbows and knees, in addition to forward flexion of the trunk. Bird[32] made the observation that joint laxity, be it joint hypolaxity or joint hyperlaxity, followed a Gaussian distribution (normal bell-shaped population curve).

Wagner and Dreschner[241] criticized the use of simple goniometers on the basis that the amount of force applied to a joint was not controlled objectively. To circumvent this problem, many attempts have been made to analyze passive joint ROM by using devices with preset torque adjustments.[33, 89, 106, 136, 200, 201, 240, 241] However, actually quantifying and documenting joint stiffness (resistance to passive movement) involves more than goniometric measurements with a preset torque. Without adequate norms to prescribe the appropriate force (preset torque), the validity of such goniometers can be seriously questioned.

Goniometric measurements of ROM with or without a preset torque as well as mobility testing of patient populations further clarify and partially document the subjective and objective measurements of joint stiffness. The actual quantification of joint stiffness however remains incomplete with these two systems of measurements.

d. Quantification of End-Feel Measurements

Previous mention has been made regarding end-feel sensations at the end of passive movement. Discussions about end-feel and its significance abound in manual therapy textbooks [59, 117, 141] and manual therapy journal articles.[40, 54, 55, 86, 172, 173] A recent textbook of goniometry[157] introduced the concept of end-feel in conjunction with ROM goniometric measurements. This inclusion indicated a new awareness of the need to record both osteokinematic joint ROM and quality of joint movement.

i) Maitland's movement diagram

Joint resistance (stiffness) in all parts of the available joint range and end-feel sensation can be documented via Maitland's movement diagrams. Maitland emphasized the necessity to realize differences, often subtle, between a "free-running" normal joint movement and an abnormal movement with minor to major resistances felt anywhere in the total ROM.[141] For the purposes of this study, joint stiffness (R), in the absence of pain and muscle spasm, is depicted. Joint stiffness itself may be due to a shortening of the connective tissues within muscles or joint capsules, scar tissue, or osteoarthritic changes in arthritic joints.[141]

Figure II-12 (A, B, C) illustrates the elements of a movement diagram. The horizontal axis AB indicates total available range of passive movement, whether osteokinematic or arthrokinematic (traction or gliding). The vertical axis AC indicates intensity of joint resistance. Thickness of the final line BD graphically depicts the sense of depth in the end-feel, whether normal or abnormal. As the joint is gently moved through the four different grades of motion, the first indication of a resistance is labelled R_1 (horizontal axis), with the end limit of total range labelled L. Vertically above L, R_2 is indicated on the CD line to identify that it is resistance or stiffness that limited the passive range.

Figures II-12A illustrates the location of the movement diagram ABCD within a force-displacement curve with its toe, linear, and plastic reions. Examples of the possible types of joint stiffness seen clinically are:

Figure II-12B: Initial resistance is felt early in the joint range; this resistance increases markedly at the end of the available range (capsular end-feel).

Figure II-12C: After initial resistance, there is a sudden limit to further movement (bony end-feel).

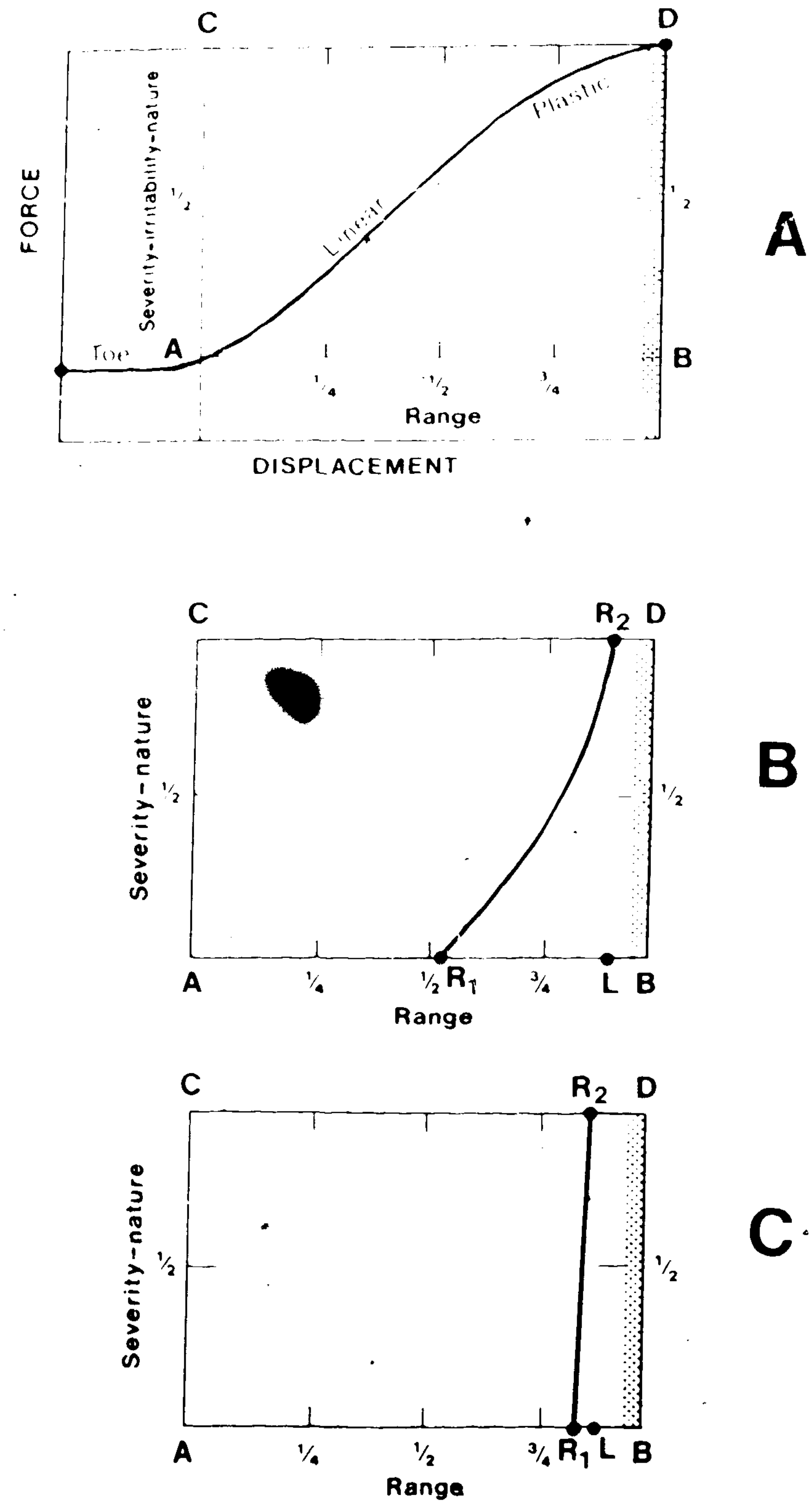


FIGURE II-12 Maitland's Movement Diagram. Details are described in the text. (Adapted from Maitland.[141]).

ii) Stoddard Paris joint mobility classification

A numerical rating scale for classifying joint motion, as observed on accessory joint play testing, was originally devised by Stoddard[209] and later revised by Paris [172] (Figure II-13). Joint hypomobility consisted of classes 0-2 with normal joint mobility as class 3 and joint hypermobility as classes 4-6. If applied to the mobility classification for population ROM testing, as described by Bird,[32] the Gaussian distribution would most likely extend from joint motion classes one through four.

However, these two systems (Stoddard-Paris and Maitland's movement diagrams) are based on the physiotherapist's ability to recognize and palpate end-feel correctly. According to Beal et al[28] and Gonnella et al[82] intratherapist reliability remained high, while intertherapist reliability proved quite inconsistent when evaluating end-feel sensations. Gonnella et al[82] pointed out that many of the measurement tools in physiotherapy such as the Stoddard-Paris joint classification were semi-quantitative.

Notwithstanding the inadequacies of such systems, rating scales can function as an "intermediate step in the development of more precise tools".[82]. The overall accuracy and reproducibility of noting end-feel on Maitland's diagrams depends on the experience of the therapist and the Stoddard-Paris rating scale only partially describes the quality or type of joint stiffness felt on palpation.

2.9 Types of Joint Stiffness

According to Ingpen and Kendall,[102] "joint stiffness affects the essential dynamics of the joint it involves, and hence the ideal measurement is one which allows free movement, is not accompanied by pain and is not controlled by the patient in any way". Wright and Johns[260] not only examined the articular physical properties that contributed to stiffness but also attempted to objectively quantify this stiffness by means of an arthrograph. The human second metacarpophalangeal joint was passively cycled through a specific ROM of flexion and extension. From the resultant hysteresis loops of torque-rotation or torque-angular displacement curves, components of joint stiffness were identified. Using

HYPOMOBILE	0 - ankylosed
	1 - marked restriction of joint mobility
	2 - slight restriction of joint mobility
NORMAL	3 - normal
HYPERMOBILE	4 - slight increase in joint mobility
	5 - marked increase in joint mobility
	6 - dislocation imminent — unstable

FIGURE II-13 Stoddard-Paris Joint Mobility Classification.
This numerical rating scale grades passive accessory joint play movements as hypomobile (Classes 0-2), normal (Class 3), and hypermobile (Classes 4-6).[172, 209]

engineering-mathematical elements in conjunction with biomechanical terminology, the authors described five different types of stiffness (elastic, inertial, viscous, plastic, and frictional).[108, 259, 260, 261] Each of these types of joint stiffness are now described

a. **Elastic Stiffness (Figure II-14A)**

Elastic stiffness, as illustrated by an ideal spring, shows a linear (Hookean) relationship between force and elongation, force and displacement and, likewise, torque and rotation. With a stiffer spring, the increase in stiffness is shown as a steeper slope on the force-elongation, force-displacement, or torque-rotation curves. Elastic stiffness, as illustrated by a rubber band, shows a non-linear (non-Hookean) relationship between force and elongation, force and displacement, or torque and rotation. A stiffer rubber band also yields a steeper slope or greater stiffness.

Whether the curve depicting elastic stiffness is linear or non-linear, the graphs present displacement (or rotation) along the horizontal axis and force (or torque) along the vertical axis. The slope of this curve is a measure of elastic stiffness (gram-centimeters/radian).

b. **Inertial Stiffness (Figure II-14B)**

Inertial stiffness, illustrated by a mass moving on frictionless bearings, shows a linear relationship between force and acceleration.

c. **Viscous Stiffness (Figure II-15A)**

Viscous stiffness, as illustrated by a dashpot-mathematical element, hydraulic cylinder, or needle syringe, shows a linear (Newtonian) relationship between force (or stress, or torque) and velocity. An increase in viscosity results in a steeper slope on the torque-velocity or force-velocity curve.[108, 245, 259, 260, 261] The slow movement of a plunger into the fluid-filled contents of a syringe requires a small force. The faster the plunging movement, the greater the resistance encountered. In a similar fashion, the engineering dashpot symbol represents a piston moving against fluid contained within a cylinder, and is the Newtonian symbol to represent viscosity.

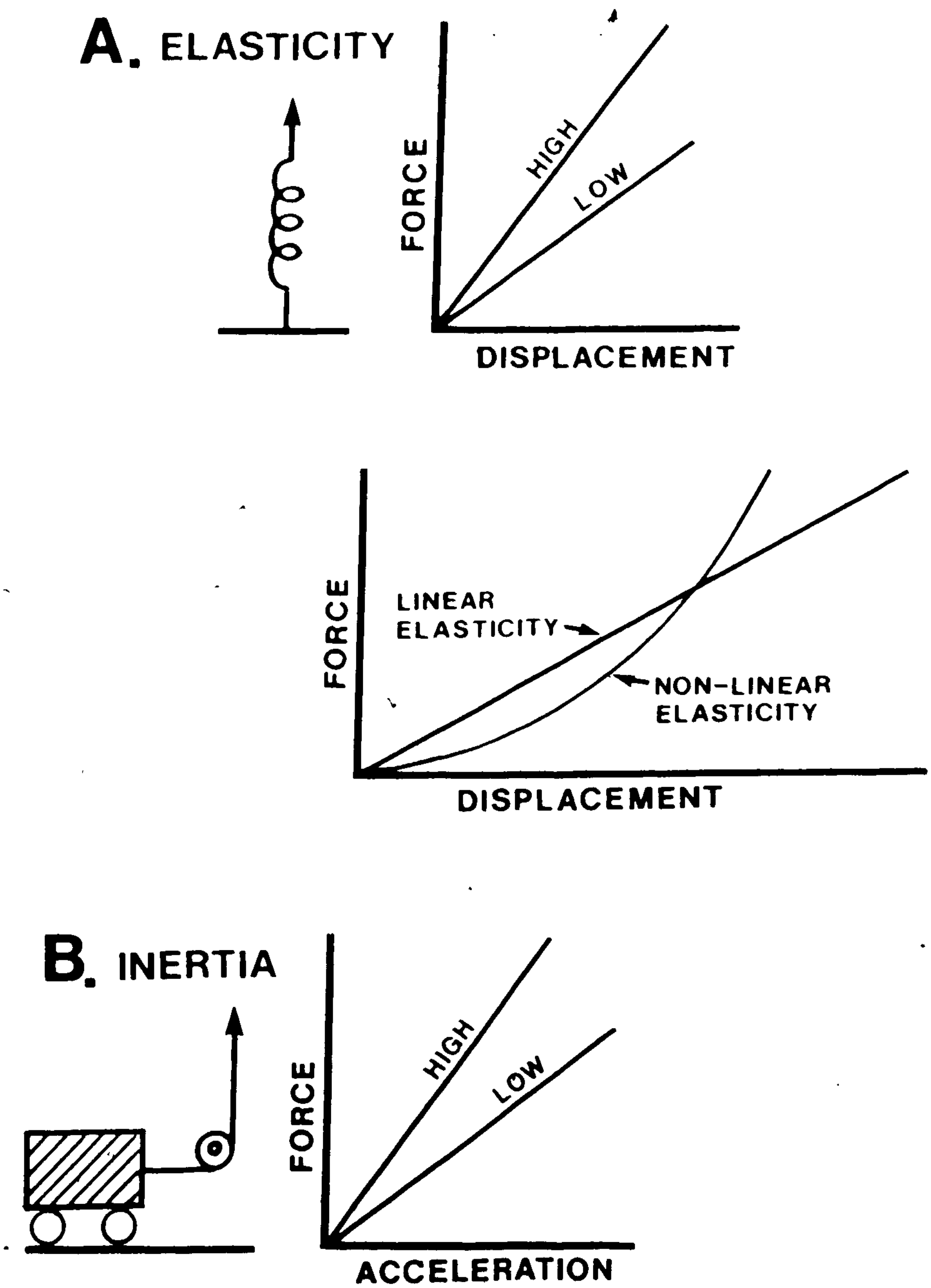


FIGURE II-14 Elastic and Inertial Stiffness. Details are described in the text. (Adapted from Wright and Johns.[260]).

A. Elastic Stiffness

B. Inertial Stiffness

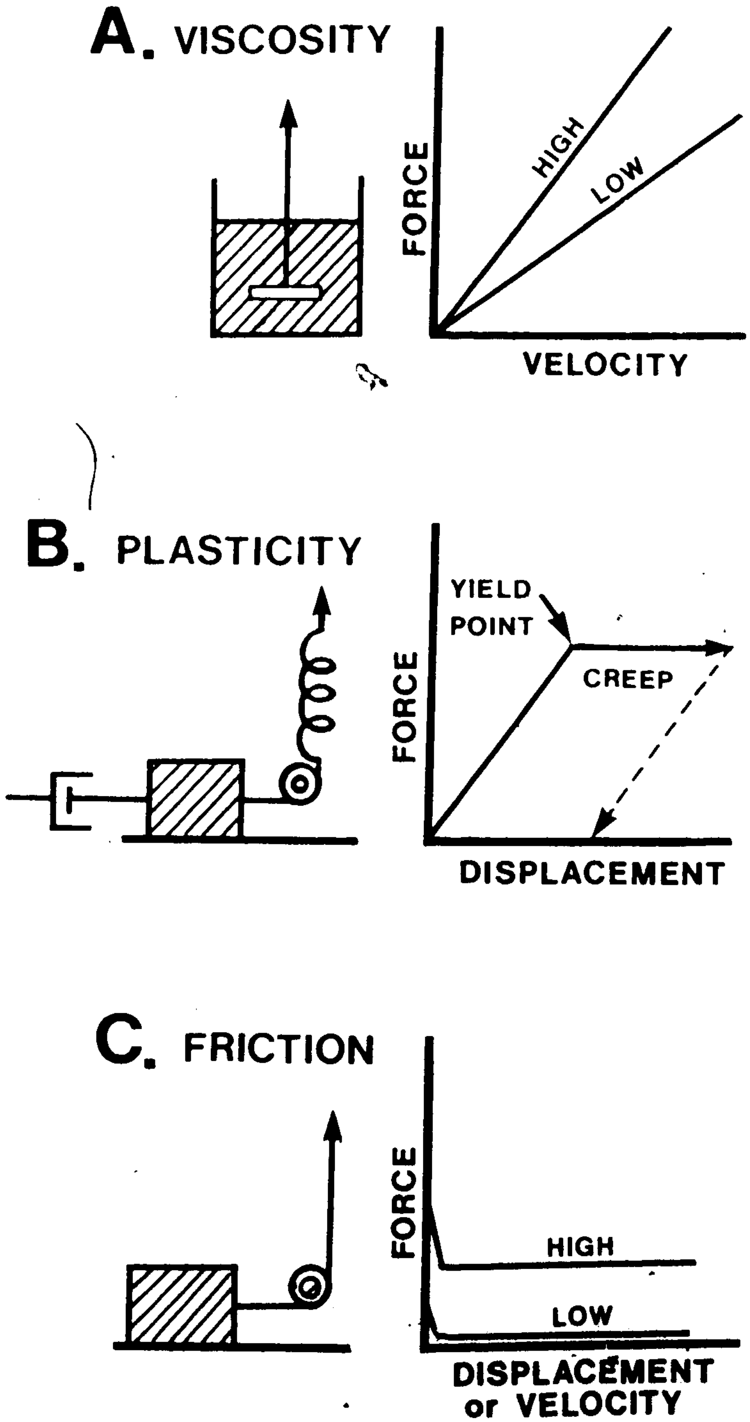


FIGURE II-15 Viscous, Plastic and Frictional Stiffness. Details are given in the text. (Adapted from Wright and Johns. 260).

A. Viscous stiffness
B Plastic stiffness
C. Frictional stiffness.

d. Plastic Stiffness (Figure II-15B)

Plastic stiffness (visco-elastic stiffness) can be explained by means of a St. Venant body and a Bingham element. A St. Venant body consists of a block with an attached spring moving over a rough surface. With an increasing force, the spring elongates. This elastic behaviour continues until the yield point. However, beyond the yield point, with further application of force, the block starts to slide. Upon release of the load, there is an elastic unloading of the spring. The block however does not return to its original position, and thus a permanent deformation is produced.[245, 261] Only non-linear elasticity is present in visco-elastic soft tissues.

Beyond the yield point, soft tissues exhibit not only plastic behaviour, but also viscous behaviour. To visualize this visco-elastic phenomenon, a Bingham element is used: a St. Venant body with a viscous element (dashpot) attached to the left of the block.

A visco-elastic substance possesses three other characteristics:

i) stress relaxation: When a plastic substance is stretched to a specific length, the required force (to maintain this particular displacement) will decrease over time. As infinite time is approached, only elastic force remains. [261]

ii) creep: When a substance is stretched by a given force, there is an initial rapid increase in length, followed by a much more gradual lengthening.

iii) incomplete strain recovery: Upon release of the force, the tissue returns to a length which equals the original length (before deformation force) plus the amount of creep. In other words, the original length is not re-attained.

e. **Frictional Stiffness (Figure II-15C)**

Frictional stiffness, illustrated by a block moving over a rough surface, shows that force or torque is independent of both velocity and displacement.

On review of various studies by Wright and Johns, [108, 109, 260, 261] it became apparent that in both normal and abnormal joints, the major components of joint stiffness were non-linear elastic and plastic stiffness, with elastic stiffness twice as great as plastic stiffness. The force required to overcome both elastic and plastic stiffness accounted for 90 per cent of the total joint stiffness.[261] Viscous stiffness accounted for only 9 per cent of total joint stiffness, while inertial stiffness was negligible.

At maximum acceleration, the torque required to overcome inertial stiffness was in the order of one hundredth of that torque needed to overcome elastic stiffness. The torque required to overcome viscous stiffness at maximum velocity was about one-tenth the torque required to overcome elastic stiffness.[260] In normal joints, there was no demonstrated frictional stiffness (independent of both velocity and rotation). Even in a hand severely damaged by rheumatoid arthritis, frictional stiffness (torque) was in the order of one-fiftieth of that needed to overcome elastic stiffness.[260]

III RESEARCH DESIGN

3.1 Introduction

The effectiveness of exercise and joint-mobilisation on the biomechanical properties of previously immobilised rabbits' knees was examined by first immobilising the left knees of thirty-three New Zealand white rabbits (Figure III-1). The knees were surgically maintained in a position of full flexion for fifty-seven days, during which time the rabbits were allowed free cage activity. At the end of this immobilisation period, the rabbits were divided into three groups of 11: Group 1 (immobilised-only), Group 2 (exercised), and Group 3 (exercised and joint-mobilised).

The immobilised-only group was sacrificed after 57 days and the hind-limbs were excised bilaterally. The biomechanical properties of both knees were tested by use of an arthrograph.

For the remaining two groups, the 57 days' immobilisation was followed by eight days of treatment involving either exercise (Group 2) or exercise with specific knee joint-mobilisation (Group 3). Subsequently, the hind limbs were excised and also tested on the arthrograph. The right lower limb in each case served as a control in all groups under study (Groups 1, 2, and 3) (Appendix B).

3.2 Subjects

Subjects for the study were thirty-three female New Zealand white rabbits, approximately 10 weeks old and weighing from 2.4 - 3.2 kilograms (kg). Prior to the initial surgery, overall mean weight for the three groups was 2.6 kg. The mean weight of Group 1 was 2.6 kg, Group 2 was 2.8 kg, and Group 3 was 2.6 kg. To avoid any mix-up between individual animals, the left ear of each rabbit was tattooed with a different number.

Until the time of surgery, the rabbits were housed at the Main Campus Animal Sciences Centre in standard rabbit cages with wire mesh flooring. Following surgery, also on the main campus, the rabbits were transported to the Ellerslie Animal Centre. There they were kept in individual pens with overall internal dimensions of 50 cm x 50 cm x 50 cm. The concrete floor was covered by wood

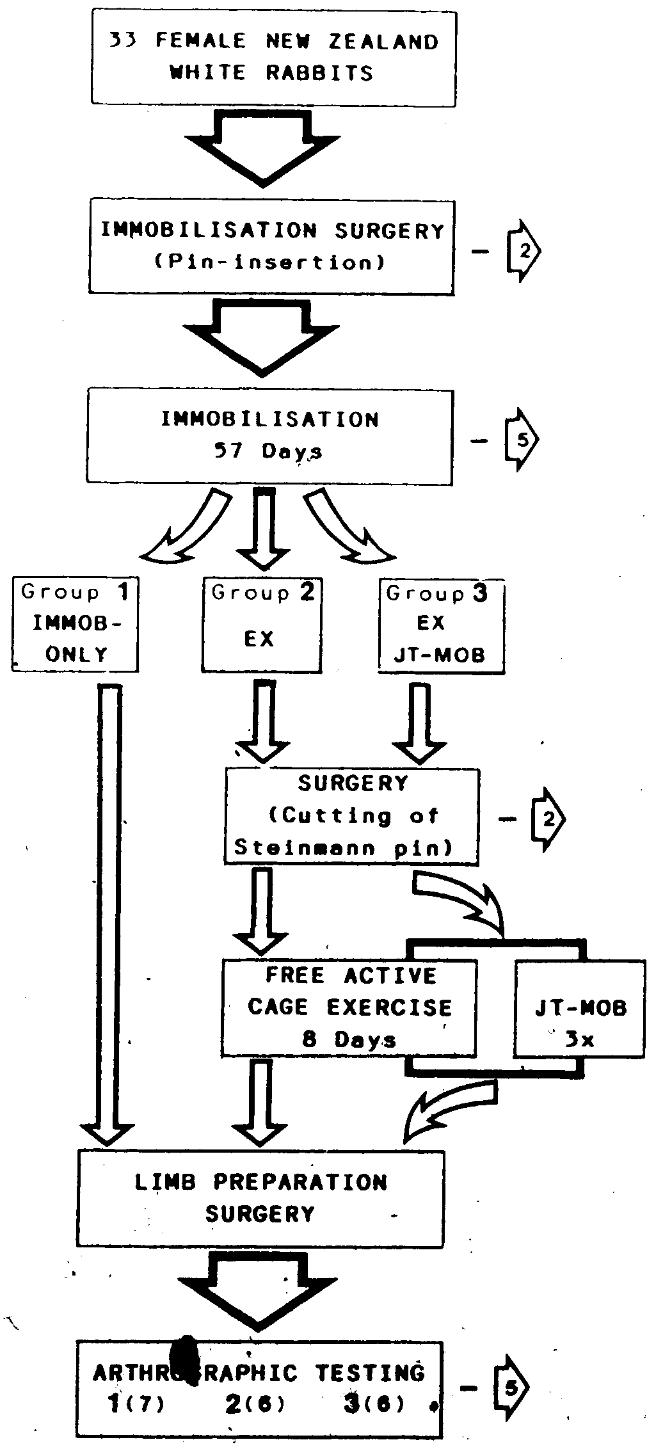


FIGURE III-1 . Flow Diagram of Research Design. Details are given in the text. The compact arrows on the right side of the figure indicate subject attrition, with the enclosed number indicating the number of subjects lost to attrition at that stage. The numbers of rabbits left for arthrographic testing were 7 in Group 1, 6 in Group 2 and 6 in Group 3.

sawdust and shavings which were changed daily. Fir plywood (G1S - 1.6 cm thick) formed the four side walls; however, the top of the pen was left open to allow a free flow of air and ensure easy access for both cleaning the pen and feeding. Food and water were provided *ad libitum*. The rabbits seemed to adapt well to their new environment despite some initial difficulty in drinking water from a bowl instead of a vertical hanging water bottle.

3.3 Arthrograph System

a. Introduction

An arthrograph was initially constructed according to the descriptions of Wright and Johns [109, 259, 260] plus Woo et al[254] and subsequently modified according to Budney (personal communication: 1985). A DC motor, with a connecting-rod linkage system, moved the tibial frame and tube (T) in an arc of motion which resulted in flexion and extension of the mounted rabbit leg. Motor speed was controlled by a potentiometer and was set at a frequency of 0.2 cps (Figure III-2).

The tibial tube frame and tube (T) rotated about axis point A of the machine in a sinusoidal motion, while the femoral tube frame (higher up) remained stationary (Figure III-3). The amplitude of this rotation, 48° to 98°, was determined by the precision potentiometer, which by design was aligned with machine axis A. The load cell, perpendicular to the tibial tube frame, recorded the torque required to flex and extend the rabbit's knee.

The femoral portion (of the rabbit's leg) was secured within the femoral tube (F), which was itself inserted into the femoral tube frame. The knee joint axis was aligned with axis A. Similarly, the tibial portion was secured within the tibial tube (T) which, in turn, was mounted inside the tibial tube frame (Figures III-4, 5, 6).

The rabbit's knee was cycled from a flexed position of 48° into the extended position of 98° and then returned to the 48° starting position (Figure III-7). This flexion-extension-flexion cycle was registered by an X-Y recorder in the form of a torque-angular displacement curve. The slope of this

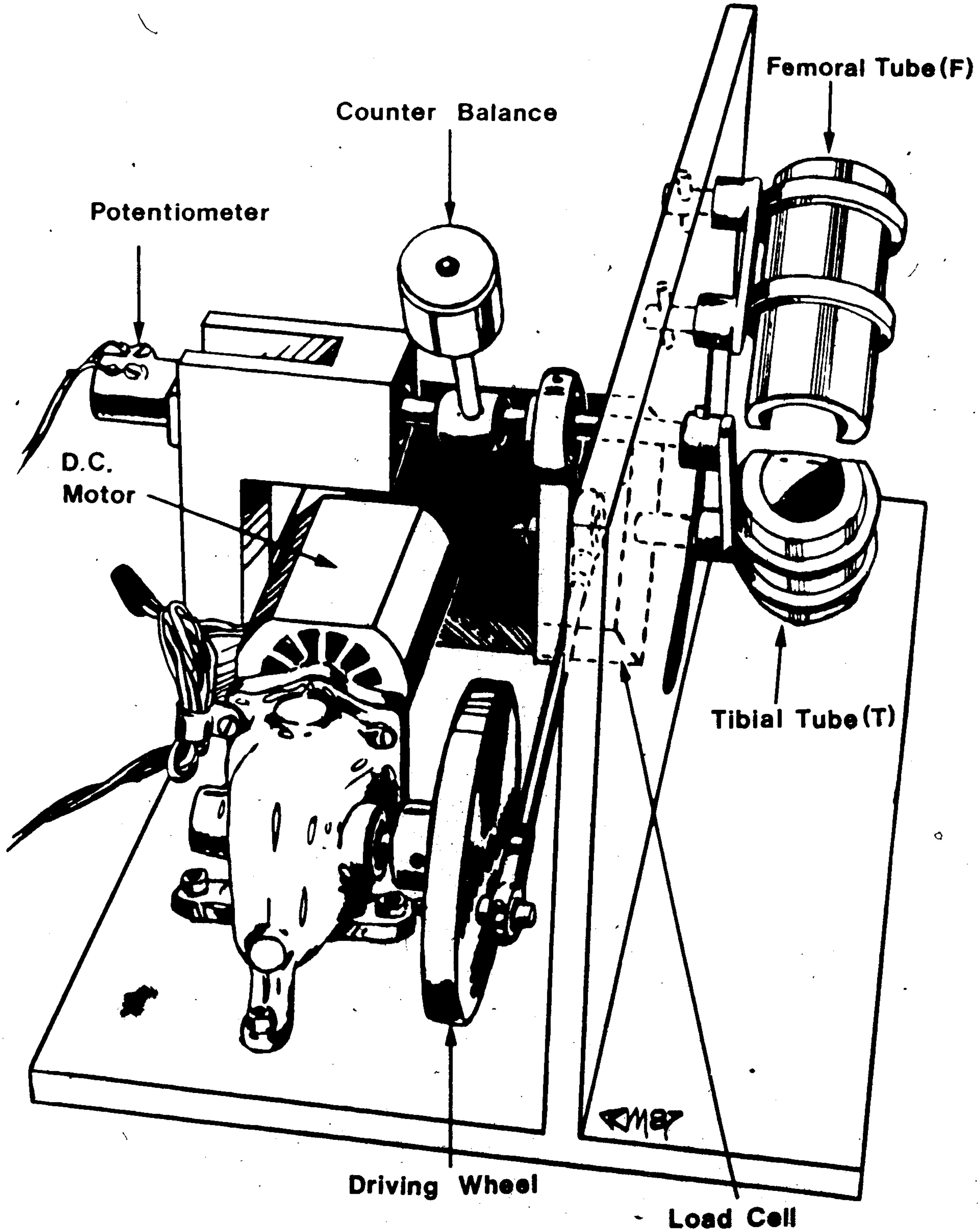


FIGURE III-2 Overall View of Arthrograph (Tubes and Tube Frames in Place). Details are given in the text.

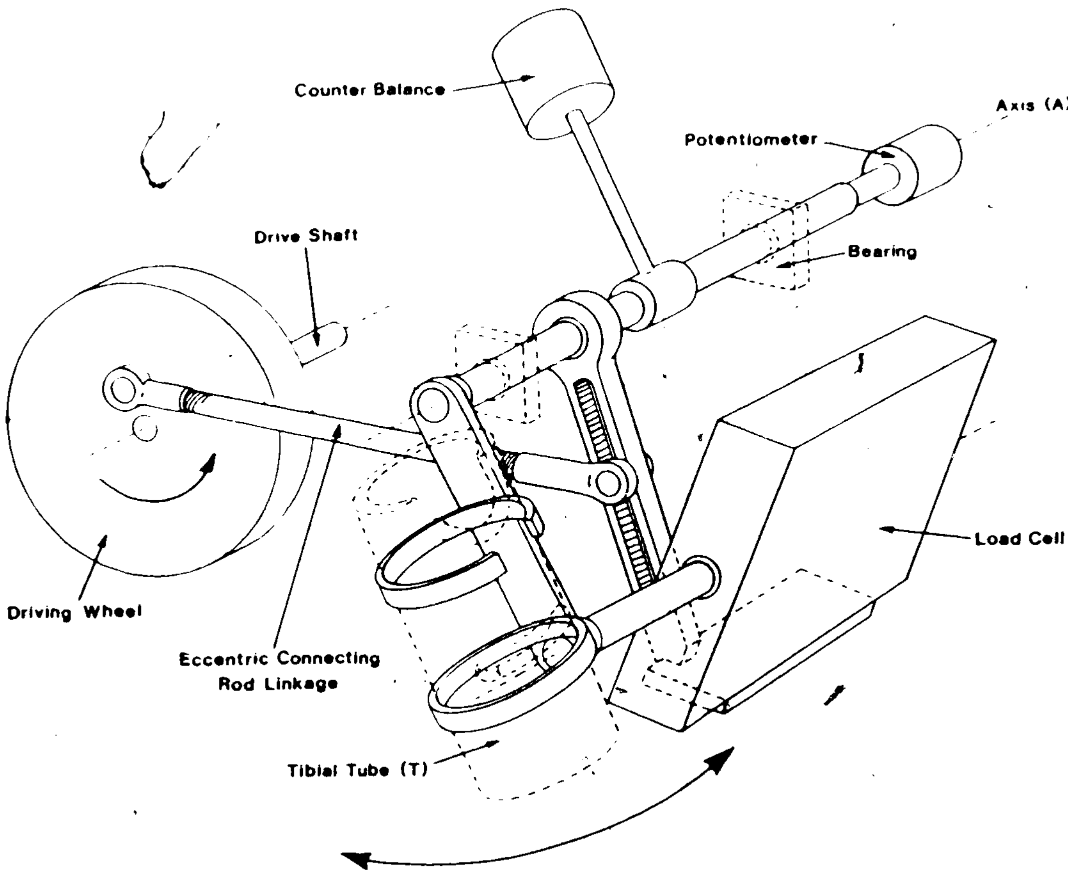


FIGURE III-3 Mechanism of Arthrograph (Chassis not shown). Details are given in the text.

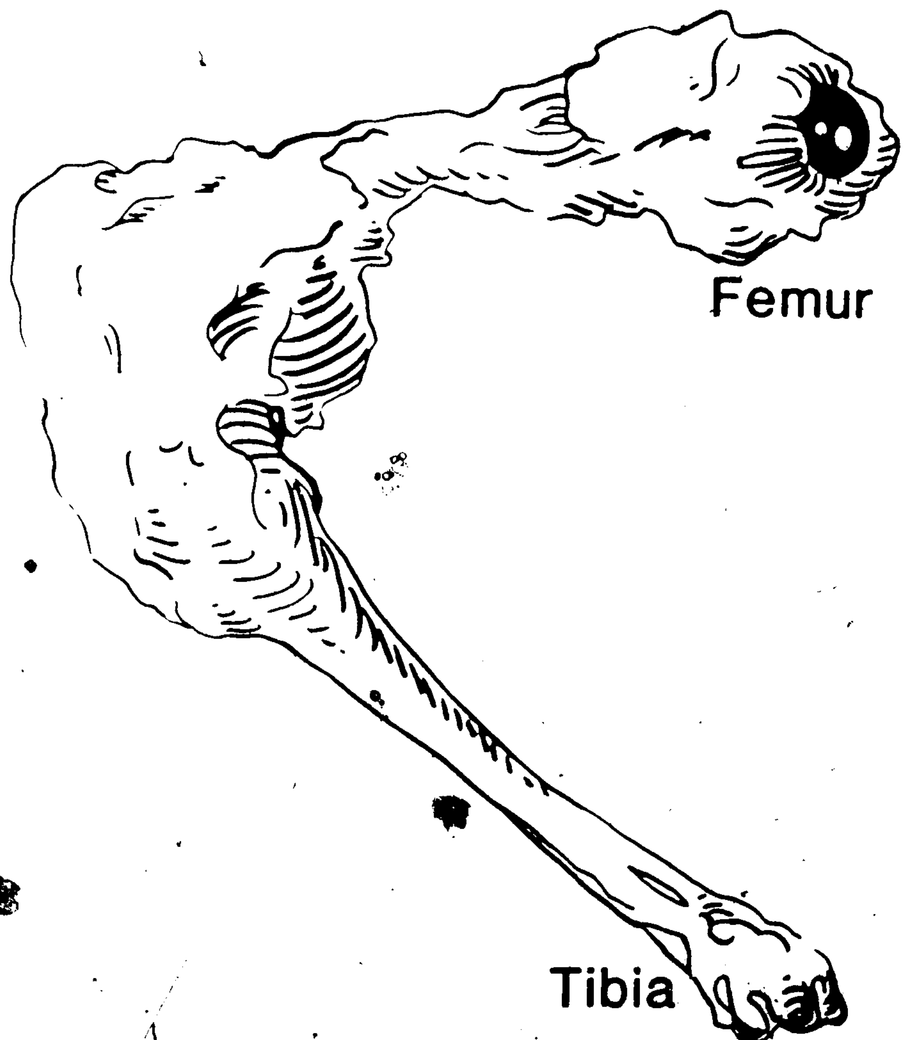


FIGURE III-4 Rabbit's Left Hind Limb, as Dissected for Arthrographic Testing.

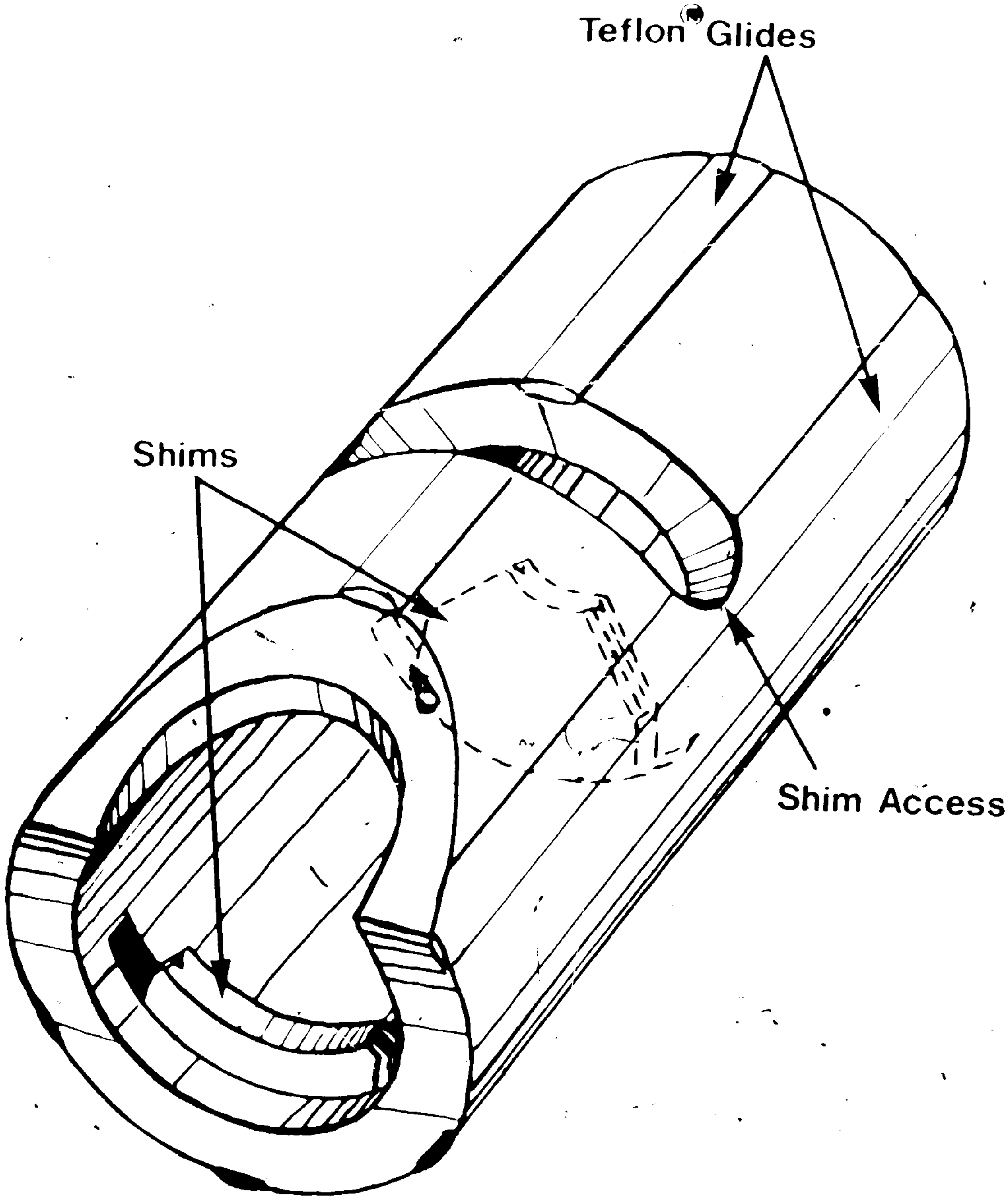


FIGURE III-5 Limb Fixation Tube (Tibial or Femoral) with Positioning Shims in Place. Teflon glides allow independent movement of the fixation tube within its own tube frame (tibial or femoral). More details are given in the text.

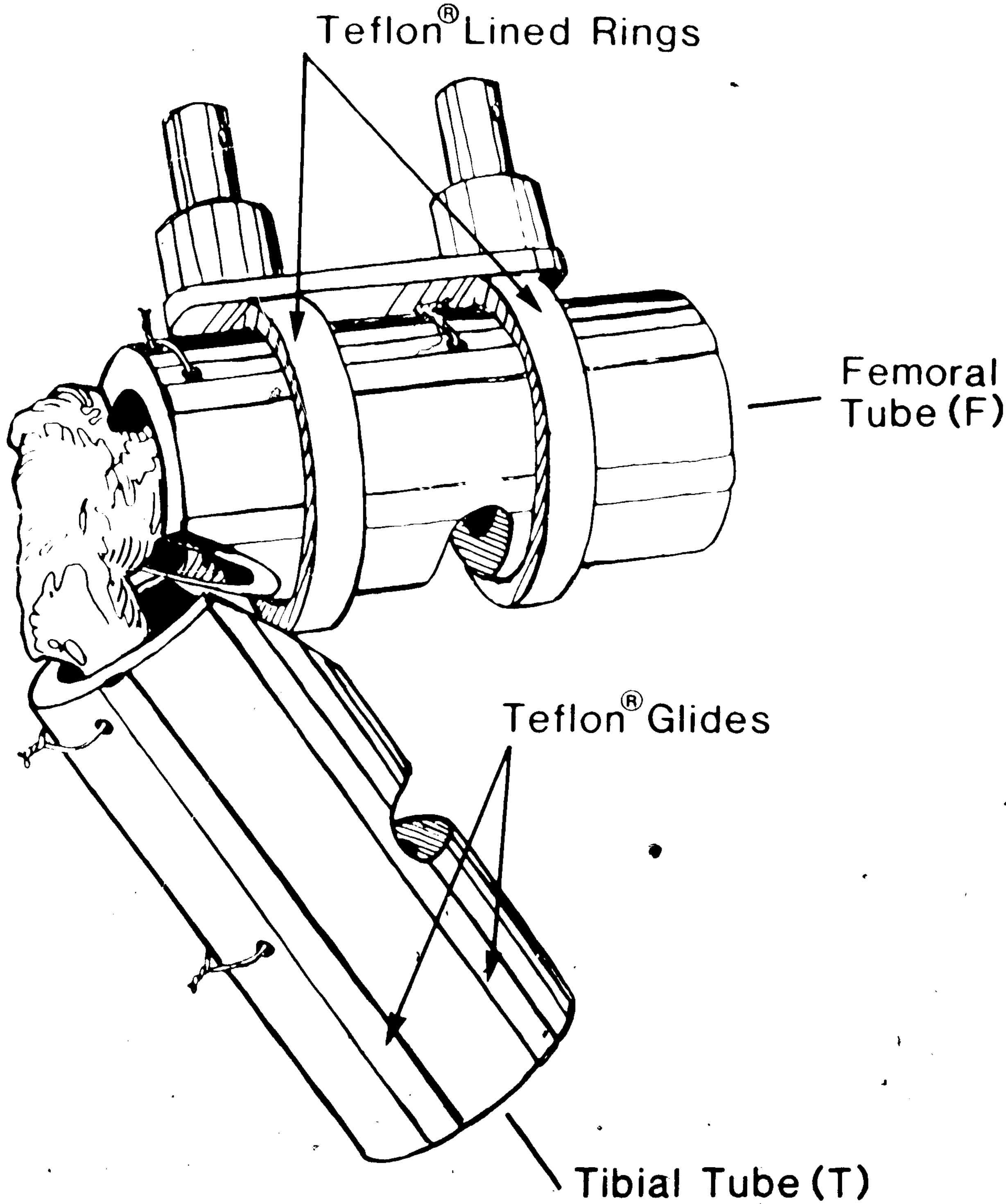


FIGURE III-6 Components of Limb Fixation System: Femoral Fixation Tube (F) in its Tube Frame, and also the Tibial Fixation Tube (T), Rabbit Limb in situ. The tibial tube frame and shims are not shown.

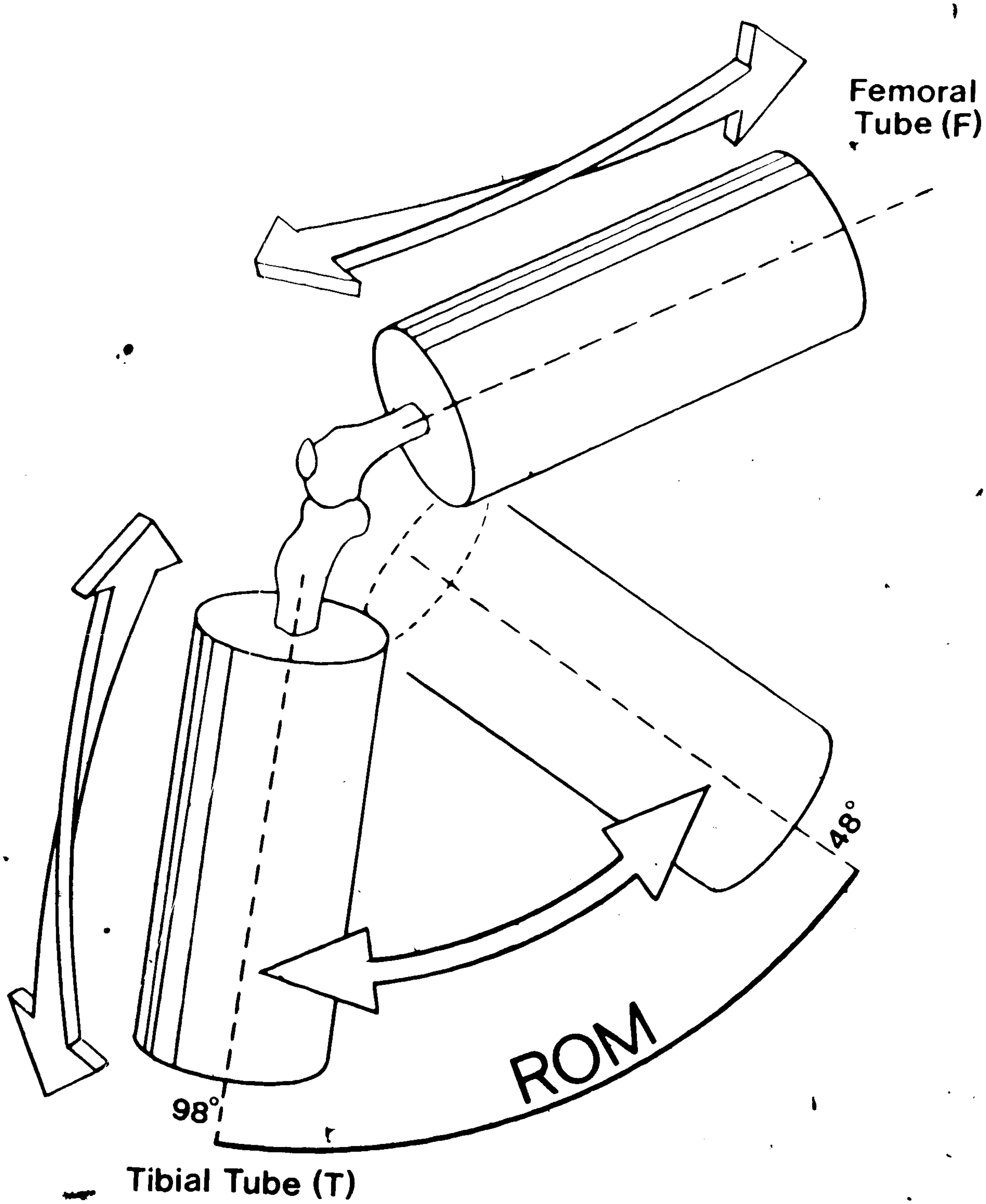


FIGURE III-7 Movement Diagram. The possible three-dimensional rotational-longitudinal movements of the limb fixation tubes within the tube frames (rabbit leg in situ) are indicated by the large twisting arrows. The flexion-extension-flexion ROM (48°-98°-48°) is indicated by the lower curved arrow and ROM.

curve was considered proportional to the stiffness of the joint.[254] The area enclosed in the hysteresis loop was considered to represent the energy needed for one cycle of motion.[254] Any resistance to this flexion-extension-flexion movement was due primarily to the knee ligaments and joint capsule, the muscle having been previously sectioned away.[254]

b. Arthrograph

The arthrograph system consisted of four separate units: i) DC power supply, ii) arthrograph, iii) load cell amplifier, and iv) X-Y recorder.

The arthrograph itself was comprised of a DC motor, gearbox, drive wheel, connecting-rod linkage system, limb fixation system, counterbalance, precision potentiometer, and load cell — all mounted on an aluminum chassis (Figures III-2,3).

The arthrograph performed three basic functions:

i) passive flexion-extension-flexion cycling of the rabbit's knee (through a specific range of motion),

ii) measurement of frictional forces (resistance forces) required to move the knee (through the arc of motion), and

iii) simultaneous recording of force (to move the knee) and angle (of the knee joint).

Range of motion was adjustable by means of connecting-rod linkages. The repeatability of a chosen arc of motion was ensured by the solid connections of these linkages. In spite of the potential for varying ROM, it was decided to keep a specific range of motion (48° - 98° - 48°) constant throughout the study. Previous researchers[6, 10, 11, 254] sequentially varied the tested range of motion from (50° - 80° - 50°) to (45° - 95° - 45°) on the same specimens. Nonetheless, it seemed to the present investigator that, by varying ROM, inconsistencies and artifacts would be introduced by setup errors in connecting-rod adjustments.

Speed of the flexion-extension-flexion cycle was controlled by an adjustable speed driving motor. Frequency of the motor speed remained at 0.2 cps or 20°/sec throughout the entire study.

The angle of the knee joint was measured by means of a potentiometer (variable resistor). This device worked by a simple electrical law $V = IR$ where V is voltage, I is current, and R is resistance. If a steady current is applied to a resistance (in this case the potentiometer), a voltage drop will occur across the resistance. If the current is maintained while resistance is changed, a proportional change in voltage drop will occur.

As a constant current passed through the potentiometer, voltage was registered by the X-Y recorder. As the axis shaft of the arthrograph rotated, the resistance changed and altered the output voltage which was documented on the X-Y recorder. The resulting signal was then fed to a load cell amplifier for amplification and demodulation. This alteration in force output was again documented on the X-Y recorder.

The principles for measuring knee joint forces were similar to those used in measuring the angle of the knee joint. A load cell or force transducer was employed using strain gauges — extremely thin, shaped wires which, by virtue of their small size, had a measurable resistance. An applied load caused a proportional change in the resistance of the strain gauge. As the wires were stretched, the resistance changed in a manner similar to that of the potentiometer, but smaller in magnitude. Because the force output of the transducer was an absolute rather than a relative value (unlike the angle transducer), the output of the transducer required calibration.

Calibration provided a voltage value for the application of a known load. Section 3.4.a describes the application of a 500 gm weight for this machine calibration. Combining the known value of the 500 gm weight with the linearity of the transducer (zero output at zero load), a correlation between load and output voltage was obtained.

Because the weight of the tibia and tube (T) provided a torque proportional to its displacement from the vertical plane through the centre of rotation, a counterbalance system was required. At the starting position of 48°, the mass of the counterbalance was to provide a torque equal and opposite to the weight of the tibia, tibial tube (T), and tibial tube frame plus, to a lesser extent, the femur and femoral tube (F).

To supplement this equal and opposite torque, an additional amount of torque (equal to the elastic resistance), was required to maintain this setup at the point of balance. However, Thompson[215] asserted that any arthrograph requiring a counterbalance could not provide absolute measurements of torque, because contributions from limb mass and elastic resistance at a joint could not be accurately separated out. If Thompson[215] is correct, then the torque measurements obtained in the present study were relative values rather than absolute values. It is unknown whether the actual relationships would change or simply the magnitudes involved. The possible problem seems to be one inherent in any mechanical design based on Woo et al's[254] design, and may itself be a subject worthy of another study.

The potentiometer used was a Fairchild (0 - 50,000 ohms) model. Ten full turns of rotation were needed to cover the entire output range of this potentiometer. Because only a small portion of that range was utilized in this application, output was easily measured with the chart recorder. Error of linearity in the output of a potentiometer is typically less than 0.25 per cent. The X-Y recorder used was a Moseley® 7005B X-Y model.

c. X-Y Recording System

An X-Y recorder yielded a torque-angular displacement curve in the form of a hysteresis loop for the flexion-extension-flexion cycle. The X-Y recorder speed selected was 0.125 sec/cm to produce the best possible graph with the chosen 0.2 cps speed of the arthrograph. The horizontal or X axis covered 25.3 cm or 50° for every cycle of flexion-extension and extension-flexion on the X-Y recorder graph paper.

Movement of the X-Y recorder pen corresponded to the straightening of the knee joint (48° - 98° flexion-extension) and was plotted on the horizontal axis (angular displacement). The torque or resistance to straightening of the knee joint motion was plotted in kilogram units on the vertical axis (vertical displacement). While the horizontal axis remained constant for all testing procedures on the arthrograph, the vertical axis varied considerably, depending on whether a treated or control knee was being tested. A sample hysteresis loop is illustrated in Figure I-1.

d. Arthrograph Leg Attachment Sites

The leg attachment sites on the arthrograph consisted of shims, a tibial tube with its tube frame, and a femoral tube with its tube frame. The femoral tube frame remained stationary, while the tibial tube frame moved through a 50 degree arc of motion in the flexion-extension-flexion cycle. The femoral tube frame could be detached from the arthrograph for ease in attaching the femoral portion of the rabbit's leg (Figure III-6). The tibial tube frame, however, was not detachable from the arthrograph.

The tibial tube (T) and femoral tube (F) each consisted of an aluminum tube with six longitudinally inlaid strips of Teflon® slightly protruding from the tube's exterior aluminum surface. These Teflon® strips contacted with Teflon® lining the metal rings (part of the tube frame) and thus allowed free movement (rotational and longitudinal) of the tube within the tube frame (Figure III-7). Each tube was fully open at both ends and had two internal slots (proximal and distal) cut into the inside wall of the tube (Figure III-5). These internal slots each extended along a 90 degree arc on the internal circumference of the tube. Three different sizes of shims fit into the slots. The sizes of shims plus position of the internal slots were determined by measurements of bone lengths, bone widths (accounting for the possibility of marked callus formation), and age range of the subject rabbits.

With correctly-sized shims in place (within the femoral and tibial tubes), the tibia and femur were each positioned as closely as possible to the centre of the tube (as viewed from the tube end) and the knee-joint axis lined up with arthrograph axis A (Figures III-5,6).

The fixation system allowed each bone to move independently of the other in response to any force placed upon it by the knee flexion-extension motion (longitudinal and rotational) (Figure III-7).

Therefore, the ability of the machine to isolate frictional forces from other forces in the knee was further enhanced.

To allow for many degrees of freedom and ensure low friction, Teflon® was used as a support between the fixed tube frames of the machine and the tubes directly supporting the leg. Teflon® strips, inlaid on the exterior tube surfaces, contacted Teflon® lining the metal rings which were part of the tube support frames (Figures III-5). The static coefficient of friction between two Teflon® surfaces is 0.04, meaning that if a weight of X units is placed on a support with both faces Teflon®-coated, the force required to start the weight moving is 0.04X.

As a result of the low coefficient of friction of Teflon®, even very small realigning forces were translated into movement which tended to realign the axis points of the knee with those of the arthrograph. The friction in the apparatus, however, was not zero. Therefore, there were some inaccuracies due to misalignment of the device. Without more data on bone characteristics, the actual error was impossible to calculate.

The arthrograph primarily measured forces applied to the knee joint itself (capsule, ligaments, articular joint surfaces) and, to a much lesser extent, elastic properties of the bones, misalignment between the knee joint and machine axis point A, and the effect of the changing angle between femur and tibia (with changing moment forces).

3.4 Calibration

a. Machine Calibration With 500 Gm. Weight

Prior to each testing session, the arthrograph system was calibrated using a 500 gm weight. With the fixed tube frames at a starting position of full flexion, the tibial tube (T) was loosely attached to the tibial tube frame with a fine wire. A 500 gm weight was then hung from the distal end of the tube.

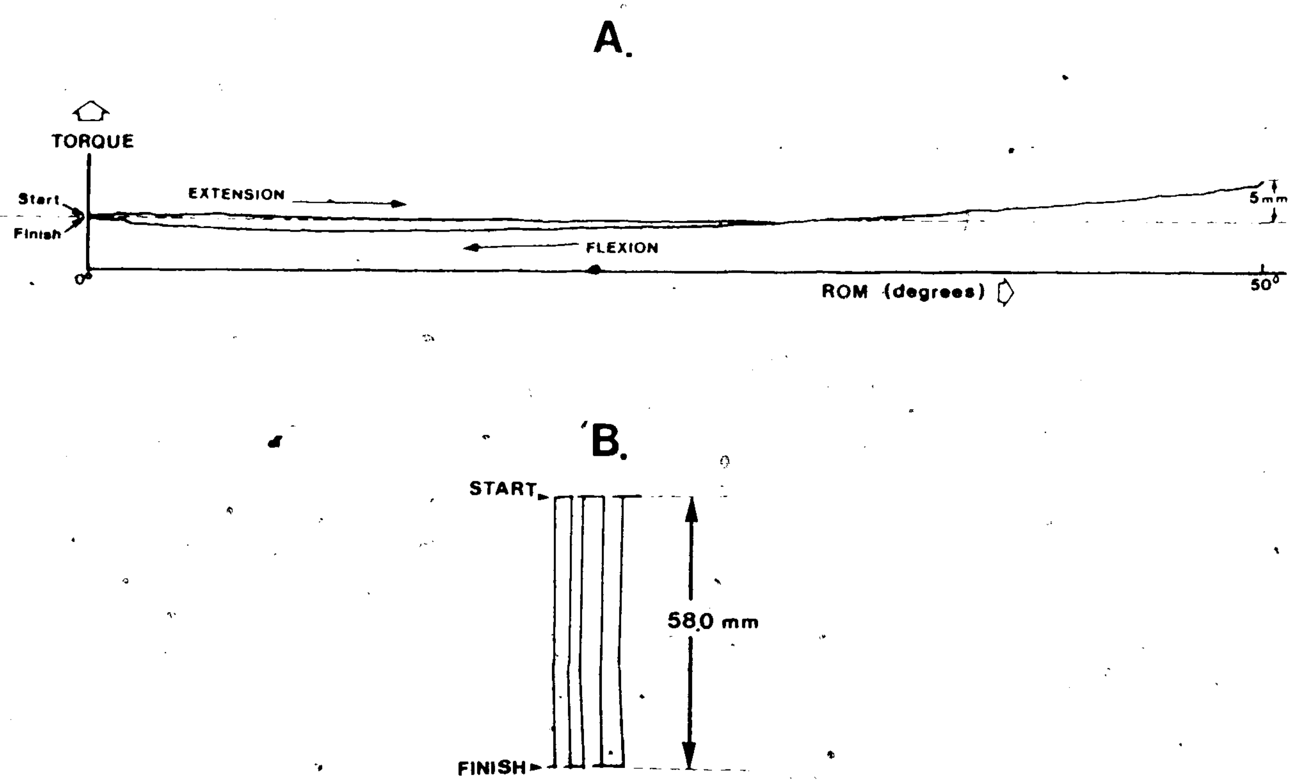


FIGURE III-8 Pre-testing of the Arthrograph Alone (Without Hind Limb in situ) for Angular and Vertical Displacement Calculations (Rabbit #21). The depicted 50° ROM corresponds to the end limits of the movement cycle 48° - 98°.

A. Machine Torque-angular Displacement Curve (Tibial Fixation Tube (T) in Place). There is a 5 mm vertical displacement near the end of the 50° cycle. This consistent machine error appeared with each machine test for each rabbit. The 50° ROM remained constant for all tests.

B. 500 gm Weight Calibration Test. Repeated vertical displacements averaged 58.0 mm. Further mathematical calculations are described in the text.

Prior to testing both the treated and control knees (Figure III-8), the amount of vertical displacement created by this weight was registered over six repetitions by the X-Y recorder. The mean average of those repetitions was used for further mathematical calculations involving the moment arm and resulted in the final calculation of vertical displacement (in newtons).

b. Machine Torque-Angular Displacement Curve

Following calibration with a 500 gm weight, the arthrograph (with tibial tube attached) was cycled through its arc of motion. The femoral tube frame remained stationary while the tibial tube frame (and its inserted tibial tube) moved in an arc about arthrograph axis A.

i) Hysteresis loops

The resultant hysteresis loops for the 1st, 2nd, 5th, and 7th cycles were recorded. The amount of resistance offered by the arthrograph system was documented before loading either the treated or control knee (Figure III-8), both in terms of enclosed area and steepness of slope.

ii) Angular displacement

Prior to testing both the treated and control knees (Figure III-8), the consistency of angular displacement was documented as the hysteresis loops for the 1st, 2nd, 5th, and 7th cycles were recorded. The angular displacement in millimeters was later converted into degrees/mm over the range 48°-98° and 98°-48°.

The use of two calibration methods (one utilizing the 500 gm weight, the other a torque-angular displacement curve) had the advantage of demonstrating that the system was functioning well as a whole.

3.5 Experimental Procedure

a. Surgical Procedures

i) *Immobilisation procedure*

The rabbit was first anaesthesized and its left lower extremity shaved from groin to ankle. This shaved leg was then sterilized and the rabbit was draped, except for its left hind limb. A sterilized sponge enclosed the left foot. Throughout surgery, the left hind limb was maintained in a position of full knee flexion.

Two incisions were made. The first, just lateral to the tibial tuberosity, separated the tibialis anterior from the tibia. The periosteum was separated medially, laterally and a little inferiorly to the tibial tuberosity.

A second incision separated the lateral aspect of the quadriceps from the tensor fascia latae. With skin and fascia held retracted from the incision site, the quadriceps was sufficiently separated from the tensor fascia latae, to allow a finger under the belly of the quadriceps to separate the middle-proximal femoral attachment of the quadriceps from the bone.

A 2.4 mm Steinmann pin was then inserted into the tibia, just below the tibial tuberosity (extending from anterior to posterior aspect). After piercing the tibia, the pin was directed into the mid-posterior femur (from posterior to anterior aspect).

A hexagonal nut was threaded onto the end of the pin that pierced the anterior femur and emerged under the quadriceps. Another nut was affixed onto the other end of the Steinmann pin located at the anterior aspect of the tibia. The knee was then brought into complete flexion by tightening the anterior tibial nut.

The various layers of tissue were stitched together, one layer at a time, with continuous sutures. Following surgery, penicillin was administered and a lateral view X-ray film was taken of the left hind limb (Plate III-1).

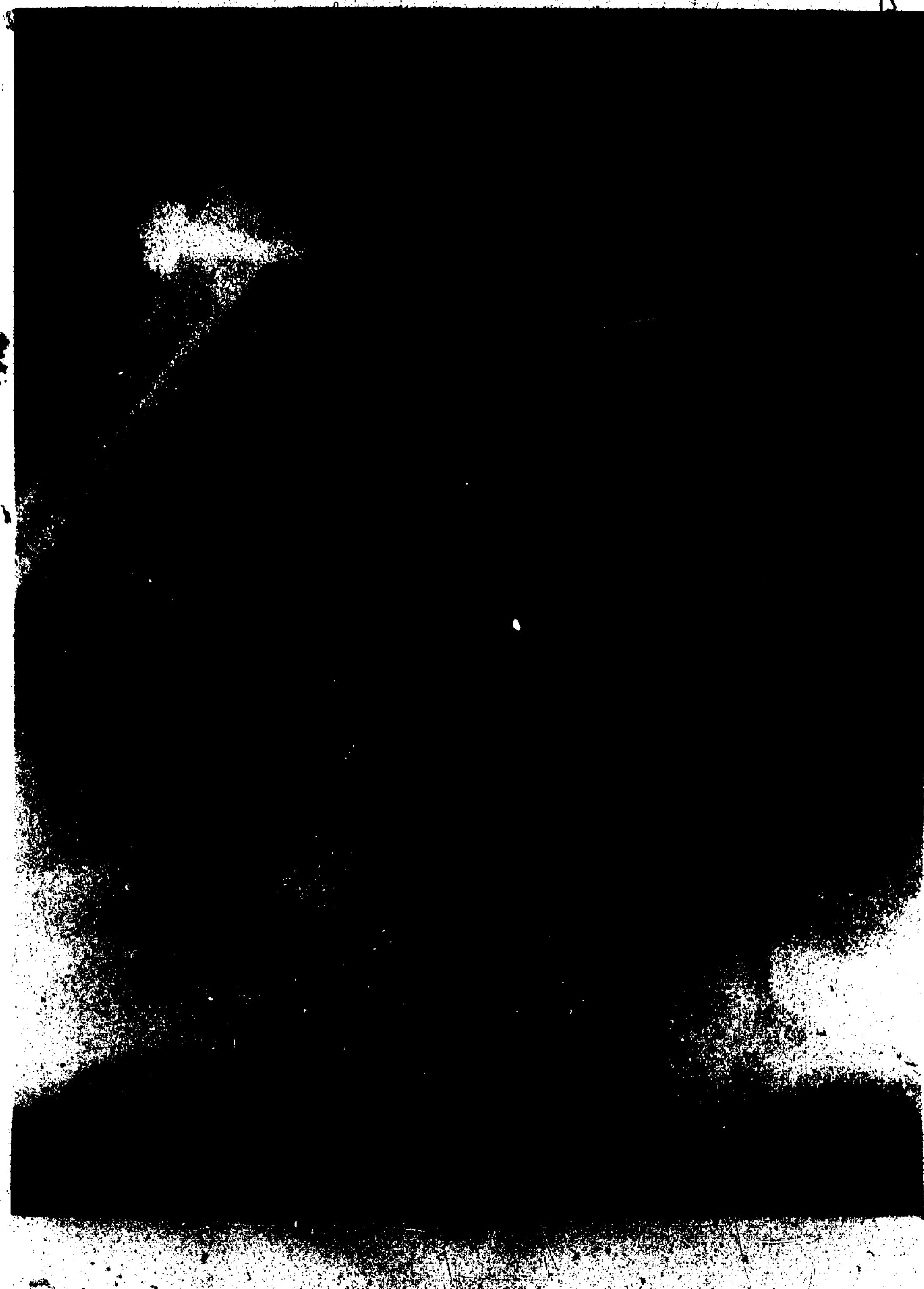


PLATE III-1 Roentgenogram of post-surgical immobilization position. Fully flexed left hind limb with Steinmann pin in situ.

The rabbits resumed normal levels of activity within 24 hours post-surgery. All rabbits moved freely in their pens, weight bearing on the left hind limb when at rest and partial weight bearing with the ankle in plantar flexion, when hopping about.

Following surgery, all three groups of rabbits were left immobilised for 57 days. Appendix D presents a more detailed description of the immobilisation surgical procedure.

ii) Cutting the Steinmann pin

After 57 days of immobilisation, both Group 2 (Ex) and Group 3 (Ex Jt-Mob) underwent a short surgical procedure to cut the Steinmann pin. The administration of anaesthetic, the shaving, and the limb preparation (skin antiseptic solution and draping) were identical to the immobilisation surgical procedure described previously.

A short lateral incision separated the quadriceps from the tensor fascia latae. Once the pin was located, the investigator stabilized the hind limb, while the surgical technician cut the Steinmann pin with wire cutting pliers. Minimal bleeding occurred. Resuturing was performed as previously described. An intramuscular injection of 0.1 cc long-acting penicillin was given just before the anaesthetic mask was removed. Subsequently, a lateral view X-ray film verified the absence of tibial and femoral fractures.

When the rabbits were alert (approximately one hour), they were transported back to Ellerslie Animal Centre. Once again, they seemed to tolerate well both the surgery and travel.

iii) Preparation for knee joint biomechanical testing

The rabbits were sacrificed by means of an intravenous overdose of pento-barbital sodium. The right and left hind limbs were excised immediately following sacrifice.

The hips were disarticulated by first extending the lateral incision between the tensor fascia latae and quadriceps (to cross over the greater trochanter), then cutting along the capsular hip joint line and, finally, severing the entire ligamentum teres. The ankles were disarticulated by cutting all tendons (dorsiflexors and plantar flexors), ankle joint ligaments, and the joint capsule.

The muscles were divided and removed in a circumferential manner with an incision (approximately 4 cm above and 3 cm below the knee joint line) just proximal to the point of pin insertion through both tibia and femur. The circumferential incision eliminated any muscle, skin, or scar involvement in the joint contracture created by immobilisation (Figure III-4).

b. Treatment Procedures

During immobilisation, all left hind limbs were maintained in a position of full knee flexion; whereas, all right hind limbs were free and acted as controls.

After 57 days of immobilisation, all rabbits were returned to the Animal Sciences Centre on main campus either for biomechanical or surgical procedures. Group 1 (immobilised-only) was sacrificed and both hind limbs were excised. Biomechanical properties of both knees were then tested on the arthrograph. Both Groups 2 (exercised) and Group 3 (exercised + joint-mobilised) underwent a short surgical procedure to cut the Steinmann pin. Both Groups 2 and 3 were allowed unlimited free active exercise in their pens for eight days. Weight bearing and use of the left hind limb were commonly observed in less than 24 hours post-surgery in both groups.

In addition to free exercise, Group 3 received physiotherapy three times for the contracture knee. Treatments commenced two days after surgery and continued on alternate days after that. The specific joint-mobilisation treatment, in sequence, consisted of:

i) 3 full passive range of knee motion exercises (flexion-extension-flexion),

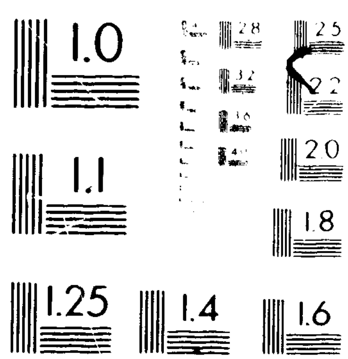
ii) 10 Grade III tibio-femoral joint traction manoeuvres,

iii) 20 Grade III anterior glides of the tibia on the stabilized femur,

iv) 10 Grade III tibio-femoral joint traction manoeuvres, and

v) 3 full passive range of knee motion exercises (flexion-extension-flexion).

2



During physiotherapy, a restraining jacket was utilized through which the left knee extended in an opening. The rabbits appeared relaxed and not in any apparent pain throughout the joint-mobilisation manoeuvres.

c. Testing Procedures

i) Specimen attachment

The femoral tube frame was detached from the arthrograph chassis and the femoral tube (F) inserted inside the frame. With the correct-sized shims in position, the femur was located with the hip joint just distal to the distal tube shim and the knee joint proximal to the proximal shim (Figure III-5). A fine copper wire was inserted through two holes on either side of the tube (forming a loop around the leg and its positioning shim), pulled up against the bone, and twist-tightened (on the exterior of the tube) to a slight tension (Figure III-6). The femoral tube frame and the tube holding the femur were then attached to the arthrograph chassis.

The tibial tube (T) was inserted into the tibial tube frame. The chosen shims, copper wires, and tibia (centred) were positioned inside the tibial tube (T). Final positioning of the tibia correctly located the knee axis position for this fully flexed starting position. The copper wires were twist-tightened (with a slight tension) on the exterior of the tibial tube (T) (Figure III-6).

When the limb was accurately centred inside both tubes and the knee joint axis aligned with axis A, the copper wires were twist-tightened to a tension holding the leg sufficiently rigid. The rabbit's leg was now ready to undergo biomechanical testing.

An identical procedure for hind limb attachment was followed for both treated and control limbs. In the legs where a Steinmann pin was inserted, the points of attachment of the pin often caused periosteal irritation which resulted in callus formation. The wide variance in size of callus made attachment of the treated legs more difficult and time-consuming than attachment of the control legs. The control legs were relatively easy to attach because the size and shape of the bones were more consistent. With its multi-sized shims and ample tube interior, the fixation system allowed all the various sized bones to be properly secured.

ii) Biomechanical testing of specimen

The procedure followed for both treated and control knees was identical. Section 3.5a iii describes specimen preparation for biomechanical testing. While one limb was being tested, the other remained wrapped in saline-soaked towelling at room temperature to prevent drying.

The 1st, 2nd, 5th, and 7th cycles of flexion-extension-flexion (through the specified arc of motion) were continuously documented by the X-Y recorder.

3.6 Overall Time Frame

All times for the three different surgical procedures (immobilisation, cutting the Steinmann pin, and sacrifice followed by specimen preparation for testing) are presented, with the exclusion of preparation and clean-up times.

The length of time for the actual immobilisation surgical procedure averaged 30 minutes. Time involved in the surgical procedure to cut the Steinmann pin was, on average, 10 to 12 minutes.

Finally, length of time to calibrate the arthrograph and biomechanically test the specimen averaged a total of 60 - 90 minutes. This total broke down into approximately 20 minutes machine calibration (2x), 20 minutes sacrifice and surgery, 10 - 15 minutes testing control knee, and 15 - 30 minutes testing the treated knee. In between tests, the X-Y recorder was turned off for a least half an hour. This proved necessary because trial testing, without a cool-off period, resulted in the production of faulty graphs. The joint-mobilisation sessions averaged 15 to 17 minutes and consisted of moving the rabbit from the pen to the treatment table (one minute), placing the rabbit inside the restraining jacket (one to two minutes), calming the subject (two to three minutes), and actually treating the subject (seven to eight minutes).

An approximate breakdown of the joint mobilisation treatments in time units identified:

i) 3 full passive ROM exercises (2x): 10 seconds x 6

ii) 10 tibio-femoral joint traction manoeuvres (2x): 10 seconds x 20

iii) 20 anterior tibial glides: 10 seconds x 20

In both cases with the tibial traction and gliding the Grade III manoeuvres took about 5 seconds to the end of the available range and a further 5 seconds to return to the initial starting position (Figures I-2, I-3).

For the final study, 15 weeks elapsed between initial surgery and the final arthrographic testing. During these 15 weeks, subject rabbits were visited five to six times per week.

Appendix E details the pilot studies undertaken.

3.7 Calculations from Torque-Angular Hysteresis Curves

a. Calculation of Moment Arm and Moment

Calculations for moment arm and moment applied during the calibration were used for *all tests* requiring this information, whether treated or control knees. Results from the machine calibration test, with the 500 gm weight, were used for making these calculations.

As depicted in Figure III-9, the 500 gm weight hung from the distal end of the tibial tube (T) where:

- tibial arm length = 100 mm
- distance 500 gm weight hung down = 28.7 mm
- angle moment arm deflected through = 9°

Moment arm (inclined at 9°) for calibration purposes:

$= 100 \cos 9° - 28.7 \sin 9°$

$= 98.77 - 4.49 = 94.28$ mm

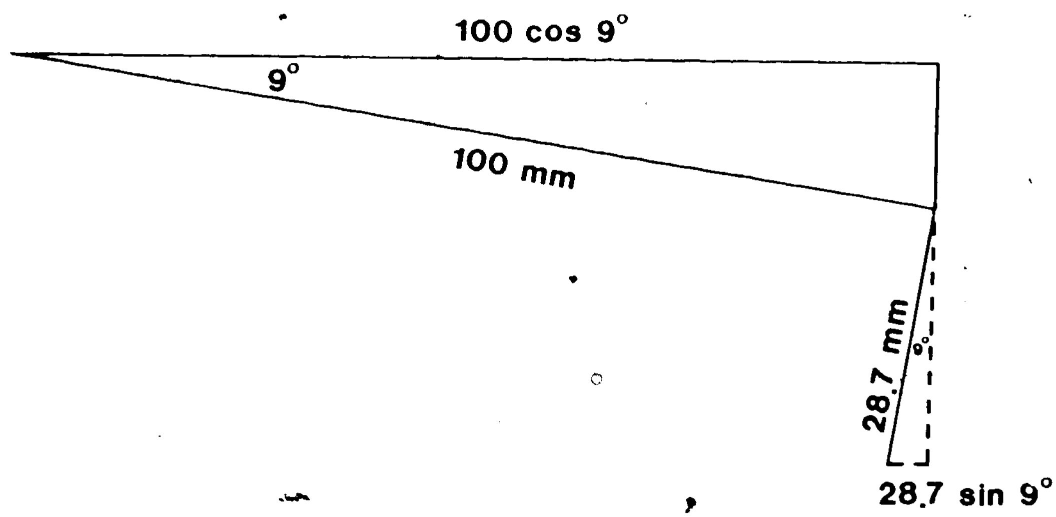


FIGURE III-9 Calculation of Moment Arm and Moment.
The 500 gm weight hangs down from the distal end of the tibial tube (T), with a tibial arm length of 100 mm, a distance of 28.7 mm that the 500 gm weight hangs down, and a 9° angle through which the moment arm is deflected. Mathematical calculations are described in the text.

If 1 kg weighs 9.81 Newtons (N)

then, 500 gm weighs 4.905 N

Moment applied during calibration:

= 4.905N x 94.28 mm

= 4.905N (500 gm weight) x 94.28 mm (moment arm)

= 462.4 Nmm

b. **Angular Displacement Along the X Axis**

The displacement range on the graph was 253 mm over the range 48° - 98° (a total of 50°).

$$\text{Therefore angular displacement} = \frac{50°}{253\ \text{mm}}$$

$$= 0.198°/\text{mm}$$

c. **Point of Linear Load**

At the point of linear load for the 1st cycle of treated knees (knee extension portion), angular displacement was read from the graph in millimeters, as was vertical displacement. An example might be the angular displacement for rabbit #33 which was 81 mm and the vertical displacement which was 111 mm (point of linear load) (Figure III-10). To convert angular displacement from millimeters to degrees, the millimeter value was multiplied by 0.198°/mm. To convert vertical displacement from millimeters to Nmm, the millimeter value was multiplied by 7.88N. A more detailed description of the calculation of vertical displacement from mm to Newtons and angular displacement from mm to degrees can be found in Appendix F.

Occasionally, there was no obvious point of linear load as described by Butler et al.[45] If that was the case, the point of maximum load at failure or the point where the graph levelled off, were used instead.

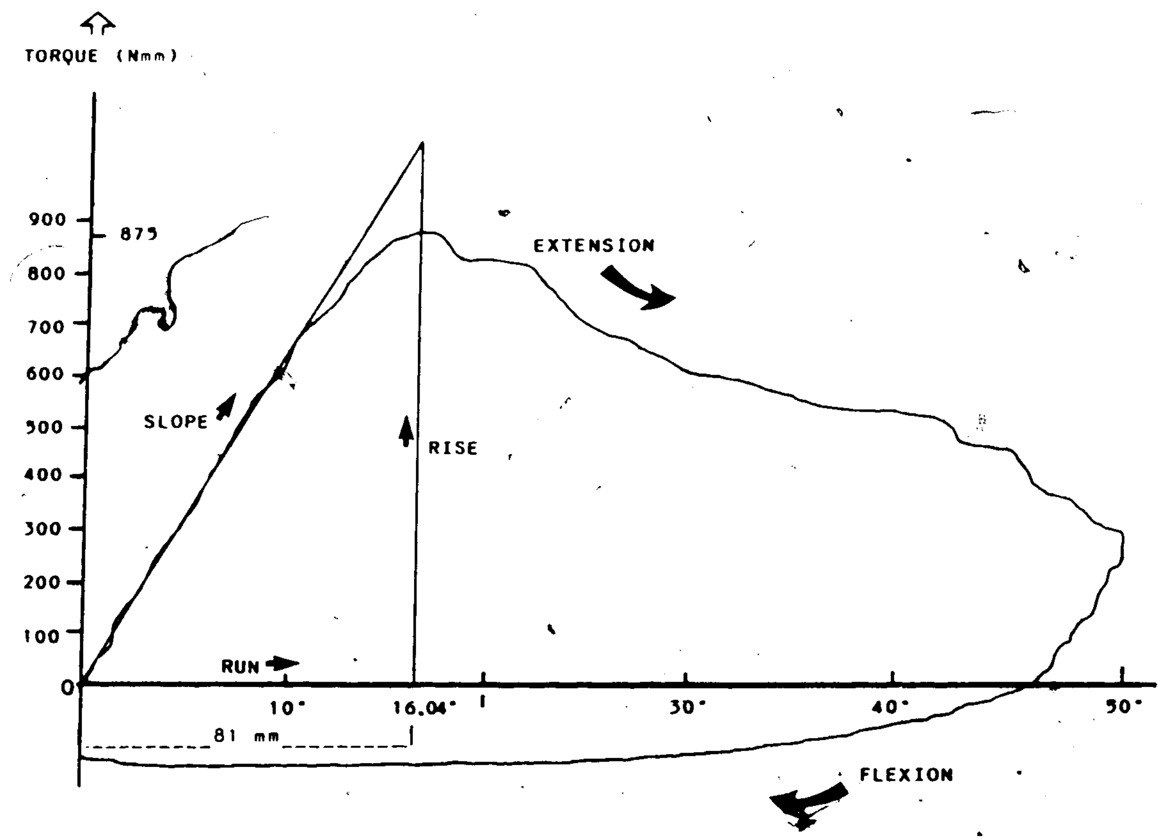


FIGURE III-10 1st Cycle Stiffness Slope Calculation to the Point of Linear Load (Rabbit #33 Immob-only). Slope is calculated from the extension curve of the rabbit's knee. Mathematical calculations are detailed in the Appendix.

d. **Mid-range and End-range Stiffness**

Both the mid-range and end-range stiffness calculations for the 2nd, 5th, and 7th cycles (treated and control knees) were determined by reading vertical displacement (knee extension portion), in millimeters, from the graphs, and multiplying by 7.88N to convert to Nmm.

Mid-range measurements were calculated from the range 73° - 85° (63 mm) and end-range stiffness from the range 92° - 98° (32 mm). Figure III-11 shows the calculations for these mid-range and end-range measurements.

e. **First Cycle Stiffness Slopes**

The slope of the 1st cycle to the point of linear load was calculated by dividing rise by run, or vertical displacement (in Newtons) divided by angular displacement (in degrees) (Figure III-10). A more detailed description of the slope for #33 immobilised knee can be found in Appendix F.

f. **Area**

The area enclosed within the hysteresis loops from the 1st, 2nd, 5th, and 7th cycles for the treated and control knees was calculated using a CalComp 9000 digitizer. The digitizer ran as a remote access device to MTS and digitized areas in terms of square millimeters (mm^2). The calculation of energy for the #33 immobilised knee can be found in Appendix F.

3.8 Ethical Considerations

The rabbits were at all times treated kindly and provided with the necessary animal care (Appendix B). Subject discomfort was minimal during the immobilisation period and treatment sessions.

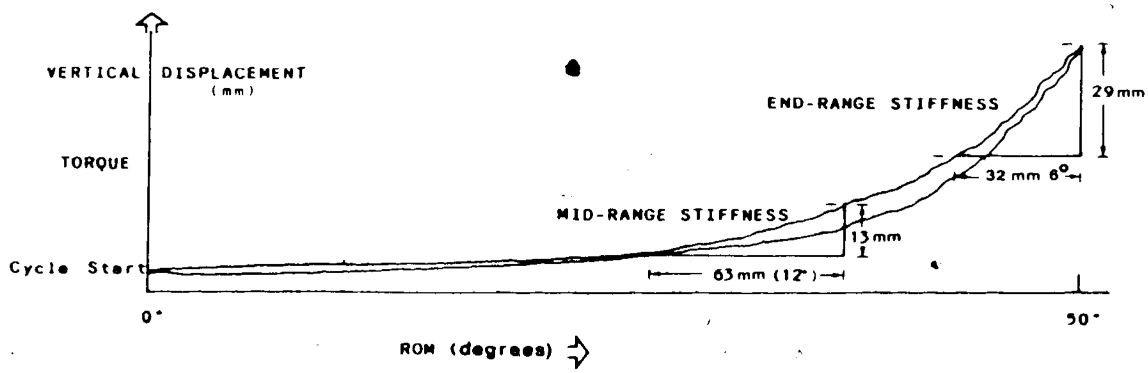


FIGURE III-11 Measurements of Mid-range and End-range Stiffness (Rabbit #39 Ex Jt-Mob) 5th Cycle. Mid-range measurements are calculated for the range 73°-85° (12° over 63 mm) and end-range stiffness for the range 92°-98° (6° over 32 mm). With a constant angular displacement for both mid-range and end-range stiffness, stiffness calculations for the 2nd, 5th, and 7th cycles are determined by reading vertical displacement for the knee extension portion in millimeters from the graphs, and multiplying by 7.88 N to convert to Nmm.

3.9 Analysis of Data

a. Interpretation

The shape of the hysteresis loop was influenced by the amount of joint contracture or joint 'stiffness' present. The hysteresis loops were analyzed in terms of the areas enclosed within the cycling loops and in terms of torque ('stiffness') measurements. Measurement of the area gave an indication of energy expended during the 50° arc of cycling motion of the rabbit's knee. The total areas of the varying cycles (1st, 2nd, and 5th) were compared. Recording of the first cycle contracture knee was particularly important because subsequent cycles required substantially less energy.

The greater the area of the hysteresis loop, the greater was the amount of energy lost during that particular loading and unloading cycle. These findings were interpreted as signifying a greater severity of the joint contracture.

The torque 'stiffness' measurements were also interpreted as signifying the greatest amount of joint stiffness or severity of the knee joint contracture with increases in the slope, vertical displacement, and mid-range and end-range stiffness. The measurements of torque or stiffness were also compared for the varying cycles of motion (1st, 2nd, 5th, and 7th).

b. Outcome Measures

The following were the dependent variables and their scale of measure:

i) areas of hysteresis (1st, 2nd, and 5th cycles) (mm^2)

ii) "stiffness" (resistance)

- point of linear load (1st cycle) Nmm
- end-range stiffness (2nd, 5th, and 7th cycles) Nmm/°
- mid-range stiffness (2nd, 5th, and 7th cycles) Nmm/°
- 1st cycle slopes (torque 1st cycle) Nmm/°

c. Statistical Procedures

The statistical technique, Analysis of Variance (ANOVA), determined whether differences among two or more means were greater than one would expect solely by chance.[96] The one-factor ANOVA examined the effect of one independent variable, while the two-factor ANOVA examined the effect of two independent variables.

For the statistical analysis comparing areas of hysteresis of treated knees for three groups over three cycles, a two-factor analysis of variance (ANOVA) with repeated measures on one factor was used:[96]

i) two factors: groups vs cycles,

ii) with repeated measures on one factor: areas as the dependent variable with three groups as one factor over three cycles as the repeated measure.

For the statistical analysis comparing first cycle vertical displacement at point of linear load of treated knees of three groups, a one-factor analysis of variance (ANOVA) was used:[96] 1st cycle vertical displacement was the dependent variable and the 3 groups was the one factor (no repeated measures).

Throughout the entire statistical analysis, a 0.05 level of significance was used and was denoted by an asterisk (*) in the ANOVA, Scheffe, and Tukey tables.

When a statistical analysis showed a significance at the 0.05 level, Scheffe comparisons of unweighted main effects, Scheffe post-hoc pairwise contrasts, and Tukey procedures were performed.[72]

Data for the torque ('stiffness') and area of hysteresis was analyzed by way of two-factor analysis of variance (ANOVA) with repeated measures on one factor for:

i) comparisons of areas (in mm^2) of control knees for three groups over three cycles;

ii) comparisons of areas (in mm^2) of treated knees for three groups over three cycles;

iii) comparisons of area ratios 1st to 2nd cycle for treated and control knees of three groups;

iv) comparisons of end-range 'stiffness' (in Nmm/°) of treated knees for three groups over three cycles;

v) comparisons of mid-range 'stiffness' (in Nmm/°) of treated knees for three groups over three cycles; and

vi) comparisons of mid-range and end-range stiffness (in Nmm/°) of treated knees for three groups over one cycle.

Data for the torque in the form of stiffness slopes and displacement values of the hysteresis loops was analyzed by way of a one-factor analysis of variance (ANOVA) for:

i) comparisons of 1st cycle stiffness slope (in Nmm/°) of treated knees for three groups;

ii) comparisons of 1st cycle vertical displacement at point of linear load (in Nmm) of treated knees for three groups;

iii) comparisons of 1st cycle angular displacement at point of linear load (in degrees) for treated knees for three groups;

iv) comparisons of 1st cycle displacement ratios vertical to angular at point of linear load for treated knees for three groups.

For the two-factor analysis of variance results, Scheffe comparisons of unweighted main effects revealed which of the trial means were significantly different from each other. For the one-factor analysis of variance results, Scheffe post-hoc pairwise contrasts of the three groups also revealed which of the trial means were significantly different from each other.

IV. RESULTS AND DISCUSSION

4.1 Attrition Rate

At commencement, this study included thirty-three female New Zealand white rabbits. This number of subjects was requisite to allow for the high attrition rate in long-term studies utilizing rabbits (Secord DC: personal communication, 1984).

At the time of surgery, two rabbits sustained fractures, one tibial and the other, femoral. Two post-operative femoral fractures occurred at four and ten days. Three weeks into the immobilisation period, two rabbits stopped ingesting food and water, while another started to gnaw at its foot (this action may have been due to nerve damage sustained in surgery) (Secord DC: personal communication, 1984). By the end of the immobilisation period, 26 rabbits remained in the study.

During a short surgical procedure to cut the Steinmann pin at the start of the treatment period, one rabbit sustained a femoral fracture while another never recovered from the anaesthetic. The total number of rabbits remaining in the study was now 24.

The testing results of a further five rabbits were excluded. During setup on the arthrograph, one femoral fracture occurred. In other cases, there were calibration or documentation problems. In one of these, the X-Y recorder overheated and incorrectly traced the knee-joint stiffness results, after the cycling had commenced. In three other cases, the 1st cycle hysteresis loops exceeded the dimensions of the graph paper. A total of nineteen rabbits remained throughout the entire study.

Groups of five or six rabbits are commonly considered acceptable by researchers (Secord DC: personal communication, 1985). In the present study, there were seven rabbits in Group 1 (immobilised-only), six in Group 2 (exercised), and six in Group 3 (exercised joint-mobilised). The Group 2 results, however, were incomplete for the 7th cycle hysteresis loop testing of one rabbit. Thus, end-range and mid-range 'stiffness' measurements over three cycles for three groups were only performed for five rabbits in Group 2.

4.2 Surgical Site

An obvious tibial and femoral periosteal external callus was created around the Steinmann pin and nuts. In addition, the Steinmann pin in the crook of the knee (posterior) was enveloped by extensive scar tissue resulting from pin-insertion surgery. Furthermore, at the nut-Steinmann pin-interface (anterior), a 'fibrous tissue capsular structure' was noted overlying the callus formation (resulting from pin insertion). This thick fibrous, pocket-like structure consisted of an inner zone of extensive fibrofatty tissue (in direct contact with tibial and femoral nuts) and an outer, thickened sheath of fibrous tissue. Both callus and fibrous tissue capsule were most developed on the anterior aspect of the femur.

The often-extensive callus formation combined with overlying 'capsular structures' sometimes prevented insertion of the hind limb into the fixation tubes. Even more importantly, it was requisite to obviate any contribution of these tissues in altering joint resistance. Consequently, the scar tissue, adjacent muscles, and overlying 'capsular structures' were all surgically removed previous to arthrographic testing.

Previous researchers, as well, have mentioned the development of 'fibrous tissue capsular structures' around internal fixation devices, both in human and canine subjects.[41] Brunet et al [41] found no impairment in the healing of bone beneath this capsule and felt that a reactive soft tissue fibrosis was occurring at the screw-plate juncture.

Past research[224] has disclosed that the extent of external callus formation seemed related to the use of metal fixation devices, but also depended upon the time period of immobilisation and subsequent remobilisation. Studies of beagle dogs involving immobilisation of the shoulder (without use of metal fixation devices) showed no evidence of periosteal callus within a six week period of immobilisation; yet during a 12 week immobilisation period, the young dogs did show evidence of periosteal reaction.

As well, these same researchers,[224] in describing the remobilisation period, emphasized the reaction of bone to a too rapid increase in overall activity level. If activity exceeded the fatigue endurance of the bone, increased intracortical and periosteal remodeling occurred.

In conjunction with an overall loss of bone strength accompanying immobilisation,[3] there was a fifty per cent loss of total torsional resistance in the diaphyseal region of the bone, caused merely by insertion or removal of a single surgical screw.[154] In animal experimentation, this reduction in strength has been reported as lasting from one to two months. Also, external callus formation was common around the insertion site of a bolt or plate.[154]

To summarize, this often extensive periosteal callus may be a result of an extensive immobilisation time period,[224] insertion of the Steinmann pin,[154] the intensive one week of remobilisation, or possibly the micromovements occurring between the Steinmann pin and bone as the rabbit hopped about in its pen (Reid DC, personal communication, 1987).

Despite development of periosteal callus and fibrous capsule surrounding the nuts, the rabbits appeared to move normally in their pens within 24 hours following both surgeries (fixation and pin cutting). Confirming this apparent comfort is Muller et al's[154] statement that an extremity with stable internal fixation was usually painless 24 hours post-surgery.

4.3 Interpretation of Hysteresis Loops

a. Shapes of Hysteresis Loops (Three Treated Groups Versus Control) From Torque-angular Displacement Curves

First cycle hysteresis loops for the three subject treated groups were basically of three different shapes, all of which showed evidence of ligamentous failure. The forms of these loops were analogous to those of load-elongation curves for ligaments elongated to failure in that:

i) Point of linear load was followed by a striking sequential failure at maximum load (Group 1) (Figure IV-1).

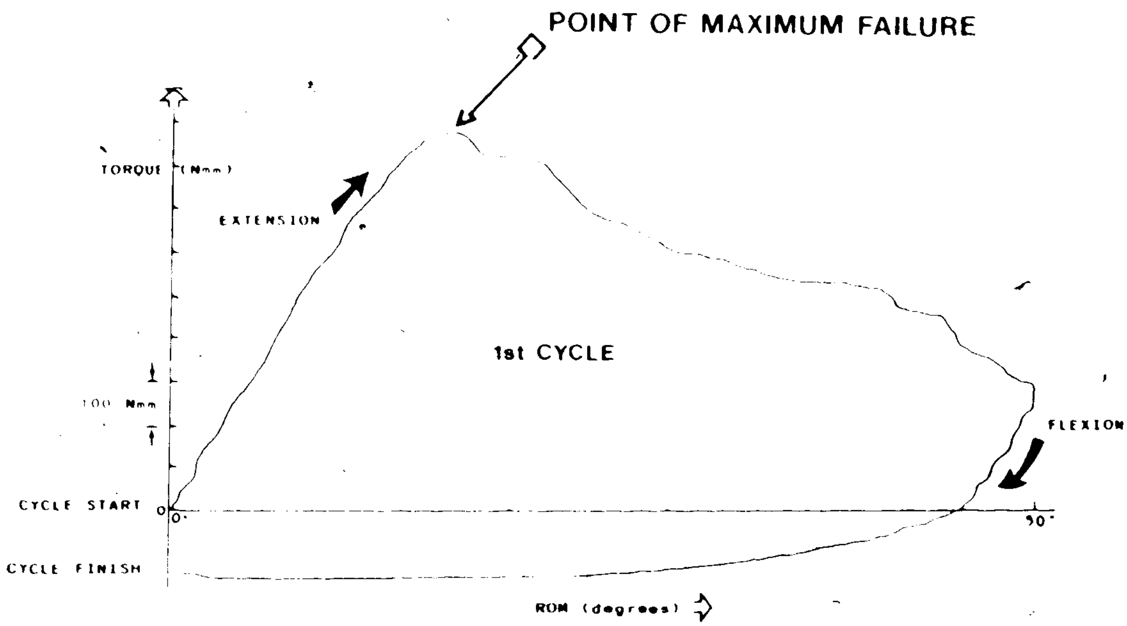


FIGURE .V-1 Torque-angular Displacement Curve (Rabbit #33 Immob-only) 1st Cycle. The point of maximum failure occurs early in the knee extension portion of the curve, following a steep initial slope. There is a marked sequential failure of the specimen to the end of the range.

ii) Attainment of point of linear load was followed by a levelling off parallel to the horizontal axis and extending to the end of the cycling motion (demonstrating elasticity and plasticity) (Groups 1, 2, and 3) (Figure IV-2)

iii) Slope slowly increased from the beginning to near the end of the total range (evidence of slight serial failure throughout the total range) (Groups 2 and 3) (Figure IV-3)

The first cycle loops (Figures IV-1, 2, 3) did not return to the starting position, an occurrence which suggested a permanent lengthening (plastic deformation). The 1st cycle loops from Woo et al's study[254] of immobilised rabbits' knees also revealed permanent deformation of the tissue at the end of the cycling ROM (Figure II-8).

In contrast, the hysteresis loops for rabbits' control non-immobilised knees revealed an extremely small loop with a minimal enclosed area as well as a minimal slope (Figure IV-4). The 1st cycle loops from Woo et al's study [254] of control knees (Figure II-8) also revealed similar hysteresis loops. For the control knees the 1st cycle loops also did not return to the starting position but were definitely closer to the starting point than were the treated knees.

To elucidate the reasons for the slopes of hysteresis loops further, it is necessary to discuss elastic plastic energy and hysteresis loops for load-elongation and torque-angular displacement curves.

b. **Plastic and Elastic Energy**

If the specimen (ligamentous or capsular) is elongated to point B (O-A-B), storing of elastic energy from elastic deformation to point B will be represented by area O-A-B-B_1-O (Figure IV-5). Within the elastic range O-A-B, portion OA represents the elastic *linear* region, while AB is the elastic *nonlinear* region. By and large, elastic energy is recoverable upon terminating the test (load-elongation or torque-angular displacement).[245]

As the specimen is loaded beyond its elastic range, plastic deformation occurs between B and C. Failure occurs at C-C_1. The total energy at failure consists of elastic energy O-A-B-B_1-O plus plastic energy B-C-C_1-B_1. Portion O-B_1-C_1 represents the permanent deformation of the specimen.[245]

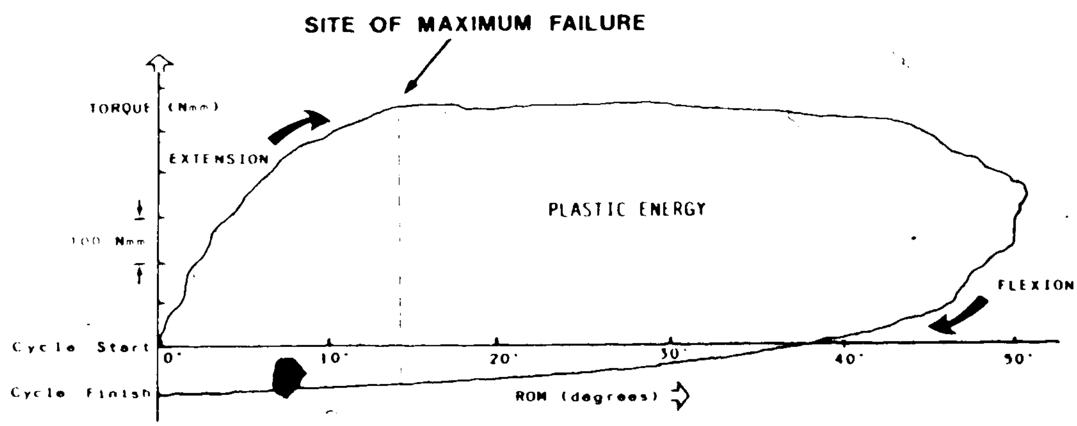


FIGURE IV-2. Torque-angular Displacement Curve (Rabbit #27 Immob-only) 1st Cycle. The point of maximum failure occurs early in the knee extension portion of the curve, following a gentle initial slope. Following failure, the curve levels off parallel to the horizontal axis to the end of the cycling motion. The region noted as plastic energy represents the permanent deformation of the specimen.

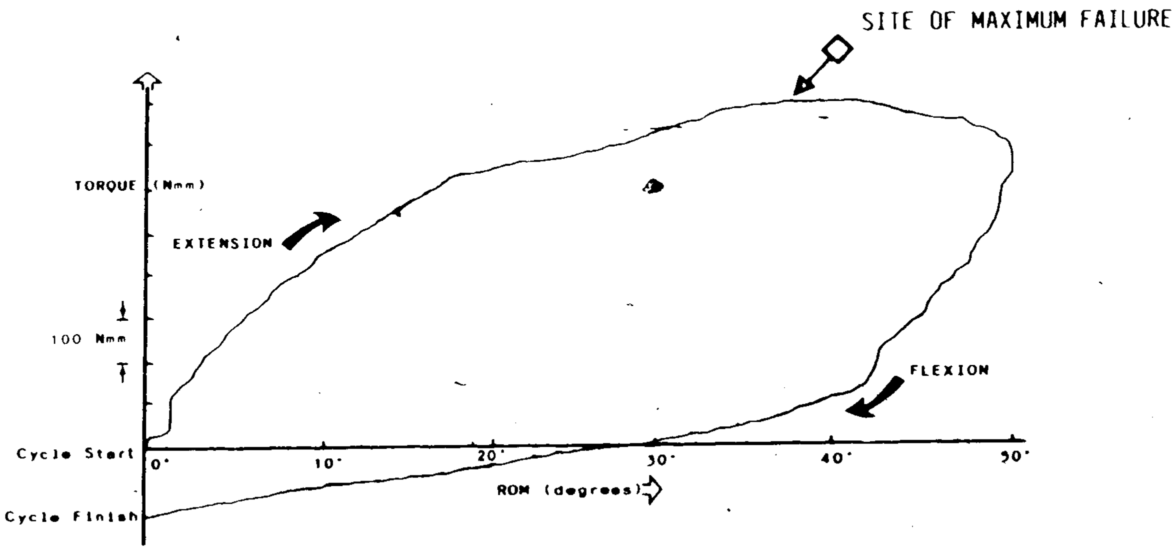


FIGURE IV-3 Torque-angular Displacement Curve (Rabbit #12 Ex) 1st Cycle. The point of maximum failure occurs late in the knee extension portion of the curve. The slope of this curve increases slowly from the beginning to near the end of the range before levelling off in the last 10°.

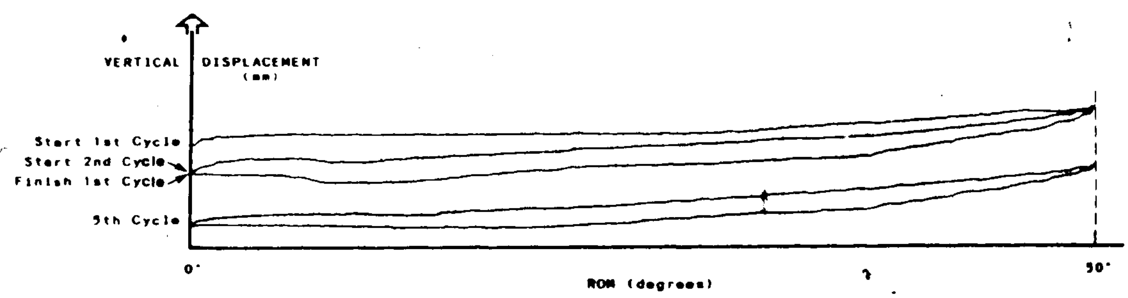


FIGURE IV-4 Torque-angular Displacement Curve for Control Knee (Rabbit #21 Immob-only) 1st, 2nd, and 5th Cycles. The minimal enclosed area of each hysteresis loop indicates a minimal loss of energy. The gentle slope of the extension curve indicates limited joint stiffness. With repeating hysteresis loops the area decreases from cycle to cycle. To convert the vertical displacement from millimeters to Nmm, the millimeter value is multiplied by 7.88 N.

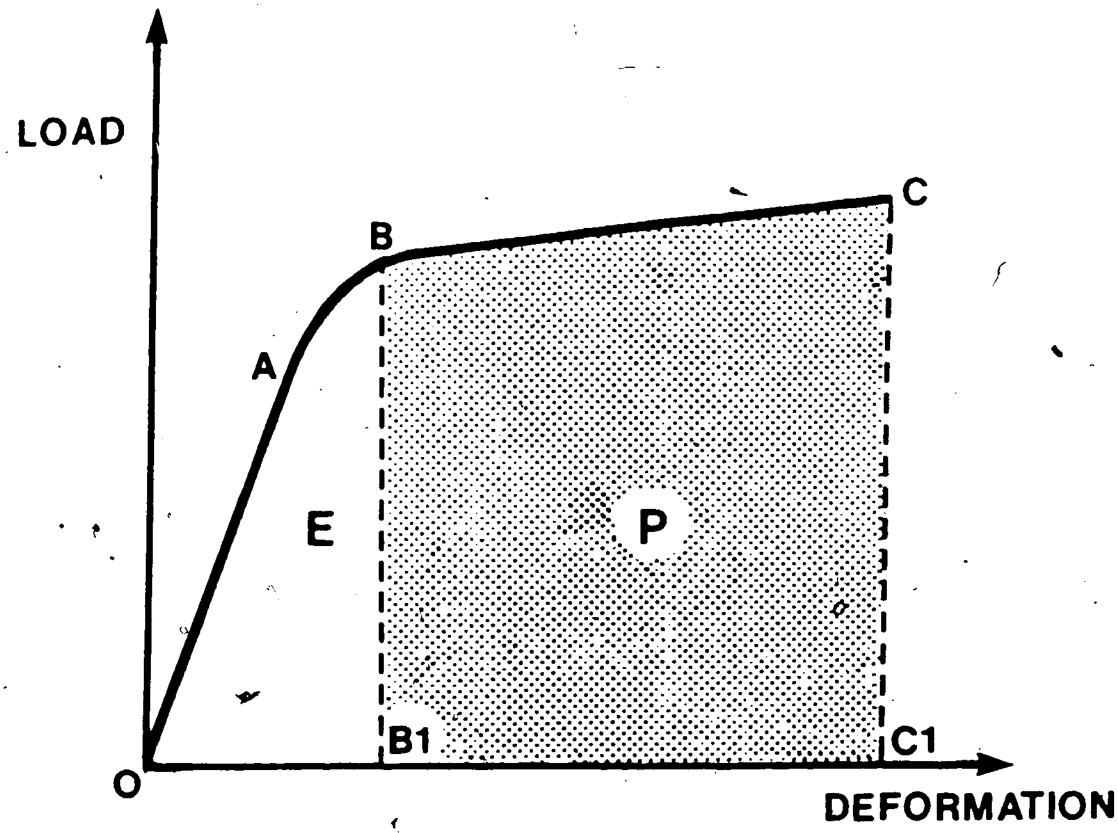


FIGURE IV-5 Schematic Load-elongation Curve with Elastic and Plastic Energy. A bone-ligament-bone specimen is elongated from O to A in the elastic linear region and from A to B in the elastic non-linear region. With further loading, plastic deformation occurs between B and C. The total energy at failure consists of elastic energy $OABB_1O$ and plastic energy BCC_1B_1. (Adapted and modified from White and Panjabi[245]).

c. **Force-elongation Behaviour of Bone-Ligament (Tendon)-Bone Specimens**

Typically, tests resulting in load-elongation hysteresis loops for bone-ligament-bone specimens are conducted in a restricted elongation range that precludes either micro-failure or macro-failures of the ligamentous tissue.[48] At a constant rate of displacement, the specimen is elongated to a predetermined peak strain (within the linear region) and then returned to its approximate initial length. The area under the loading curve represents the energy of deformation, or energy supplied to the tissue. The area under the lower curve represents energy released by the tissue during this unloading. If the unloading curve duplicates that of the loading curve, as may be with a Hookean substance, the energy of deformation is totally regained. With true viscoelastic tissues, the area of the hysteresis loop represents the energy lost by the tested specimen.[45, 245]

Hysteresis loops have only been described in a load-elongation test for normal ligamentous tissue. Load-elongation curves for previously-immobilised ligaments (as compared to controls) show: i) a decrease in maximum load at failure, ii) a decrease in total energy absorbed at failure, iii) a decrease in the linear region slope (decreased stiffness), and iv) an increase in ligamentous elongation per unit load (increase in extensibility).[45, 48, 162, 166] The hysteresis loop for an immobilised specimen would likely show similar findings with a decrease in stiffness of the linear region, a decrease in energy loss during cycling motion, and an increase in length before reaching a predetermined strain (Figure II-10).

In control ligaments, a repeated cycling to the same peak strain yielded a reduced area of hysteresis loop with each repeated loading-unloading cycle (Figure II-7). For immobilised tissue, this area may be even more rapidly reduced due to its already increased extensibility.

Tensile loading of bone-ligament-bone specimens, extended to failure, produced a typical load-elongation curve with toe region, linear region, and linear loading end-point, with its first evidence of major failure in a few overstretched ligamentous fibres (Figure I-4). Further elongation, requiring progressively less and less force, ultimately resulted in point of maximum load, directly followed by a rapid failure of the entire ligamentous structure.[45, 48, 160]

The results of this study however showed hysteresis loops extending into failure with marked serial failure (Figure IV-1) as well as gradual evidence of failure (Figures IV-2, IV-3). Both the terminology of and interpretation of load-elongation curves was required for understanding the results of the torque-angular displacement curves of this study.

Grood et al, as quoted by Butler et al,[45] studied load-elongation patterns for bone-ligament-bone specimens of the rhesus monkey's medial collateral ligament and medial capsule of the knee (Figure II-6). The medial collateral ligament reached a much higher maximum force, followed by a markedly more abrupt failure than for the capsule. At the point of complete failure of the collateral ligament, the entire capsule was supporting only half the total ligamentous force. Further elongation of the specimen produced a marked 'serial' failure of the capsule.

Serial failure for both shoulder and elbow joint capsules in the human has been described previously.[118] The collagenous fibre bundles of the medial collateral ligament tend to be more parallel and uniform in length than fibres of the anterior cruciate ligament. As a result, medial collateral ligamentous fibres tend to fail all at once rather than serially (Figure II-6). By contrast, cruciate ligamentous fibres tend to fail more sequentially (Figure I-4). And, as previously stated, capsular fibres (least organized with varying lengths and orientations) tended to fail sequentially, over a greater elongation range than for either collateral or cruciate ligaments.[45, 48, 160, 162]

d. Torque-angular Displacement Curve

The hysteresis loops from a torque-angular displacement curve for rabbits' control knees presented with an extremely small loop area in the presence of minimal stiffness (whether initial, mid-range, or end-range) (Figure II-8).[254] Earlier research involving arthrographic testing of human finger joints also revealed small areas of loop enclosure and minimal stiffness.[20, 260, 261] Force-elongation tests apply ideally to uniaxial tissues, such as collateral ligaments, but are less than adequate when applied to capsular fibres which are predominantly tested via torque-angular tests.

Cycling a previously-immobilised rabbit's knee joint through a predetermined ROM revealed a notable increase in both enclosed area and steepness of the slope (Figure II-8).[254] Similar findings, but of a lesser magnitude, were found in reference to human osteoarthritic and rheumatoid arthritic finger joints.[20, 221, 261]

The hysteresis loop from load-elongation tests of control knees showed a sharp increase of slope (stiffness) in the linear region and some energy loss (shown in the enclosed hysteresis area) (Figure II-10). The amount of energy lost in this situation does not appear to have been clearly quantified in the literature, but would likely vary according to the individual specimen and strain rate. In contrast, a hysteresis loop from the torque-angular displacement of a control joint revealed minimal stiffness and minimal enclosed area (Figure II-8).

These two types of hysteresis loops—one showing an increase in stiffness, the other a decrease — seem to be representative of the two different types of tissue. Ligamentous tissues seem more ideally loaded in tensile stress directions due to the parallel arrangements of their collagenous fibres. Capsular fibres seem much more multi-axial, with lines of force changing continually as different regions of the joint range are entered. If the nylon hose weave[3] (criss-crossing pattern of collagenous fibres) is actually present, the fibres should orient along the directions of the stress and elongate. Further into the range, other fibres become oriented to the new direction (Figure II-4).

Hysteresis loops from previously immobilised rabbits' knees produced a marked increase in both the stiffness (slope) and area within the loop.[254] Akeson et al[3] have explained this behaviour, apparently opposite to the load-elongation curve, as resulting from orientation of capsular fibres, laying down of extra collagenous cross links, gross adhesions of articular and periarticular soft tissues, and a decreased ease of gliding at articular joint surfaces.

4.4 Choice of Cycling Range of Motion

The ROM (50° arc of cycling motion) for biomechanical testing of the rabbits' knees was chosen as a result of examining the results of Woo et al[254] and Akeson et al.[11] The present investigation did not precondition the specimen as done by Woo et al,[254] but merely cycled the knee through a 50 degree arc of motion.

Butler et al[48] reported on the importance of preconditioning or cyclically elongating the ligament or tendon to no more than 20 to 30 per cent of the rupture force, in order to eliminate the 'first time' behaviour of this tissue. He further stated that certain components of the ligament "ruptured and played no part in further tests."[48] However, it was not desired to destroy the specimen in this study.

Cycles subsequent to the second loading-unloading cycle were traced on unused, progressively lower portions of the graph paper (Figure I-1). This was done to prevent cycles from tracing one over the other; however, it also meant that the successive gradual shift of the hysteresis loop could not be documented (Figure II-7). Nonetheless, it seemed more important to eliminate the interference of nearly-overlapping cycles (Figure II-8) and have cycle tracings separate enough, one from the other, to enable accurate analysis of torque and area of hysteresis.

For the immobilised-only specimens, the mid-range and end-range 'stiffness' measurements were not done due to early failure of the specimen. Only 1st cycle stiffness measurements could be done.

The initial study of Woo et al[254] involved nine weeks of immobilisation of the rabbit's left hind limb. Biomechanical testing of the rabbit's knee was first performed for the range 50 to 80 degrees and subsequently for 45 to 95 degrees. As stated in the previous section, hysteresis loops are normally tested at strain levels well below micro-failure or macro-failure of the ligamentous specimen. The lack of stated evidence of failure in studies by Woo et al[254] and Akeson et al[5] suggested that a lengthening of collagenous fibres occurred during the 1st cycle of 50 to 80 degrees (5 cycles total). Possibly, during this ROM, the ligamentous fibres were stretched or partially torn, fibres of the posterior knee capsule were stretched, and fibrous connective tissue adhesions within the joint space were stretched and torn. If so, cycling in the larger ROM (45° to 95°) would meet with less resistance.

On closer examination of hysteresis loops from Woo et al's studies,[254] it was noted that the upper curve represented the flexion portion of the cycling motion. If the graph was positioned so as to have the upper curve as the extension portion of the cycling motion, it was noted that evidence of ligamentous failure took place in the last 7 to 8 degrees. During this specific range, the loading curve flattened, levelling off parallel to the horizontal axis (Figure IV-6).

Even in the 30° cycle ROM there was evidence of ligamentous failure,[254] though not as obvious with the 50° cycle, as in the present study. Unfortunately this failure region was not noted by previous researchers.[11, 254] Subsequently, the incorrect ROM was chosen by the present investigator, since the 30° cycle would have been far superior to the 50° cycle.

4.5 Choice of Arthrograph

The arthrograph, constructed according to the descriptions of Wright and Johns[259, 260, 261] plus Woo et al,[254] and subsequently modified according to Budney (personal communication: 1985), was used for all biomechanical testing of knees from each of the treatment groups. It was hoped that with the use of an arthrograph similar to that described by Woo et al,[254] previous research findings on the effects of joint-immobilisation and remobilisation would be confirmed and the new dimension of joint-mobilisation added.

Regrettably, some basic omissions must be noted in previous arthrographic research studies (involving rabbits' knees). Throughout, no operational definitions of hysteresis and torque were given. Moreover, no information was given in any of these reference studies[6, 8, 10, 11, 16, 254] or review articles[7, 8, 12] detailing how or where in the available range, torque was measured. Furthermore, only standard deviations and means were given, without any reporting of further statistical analysis that could indicate statistically significant relationships.

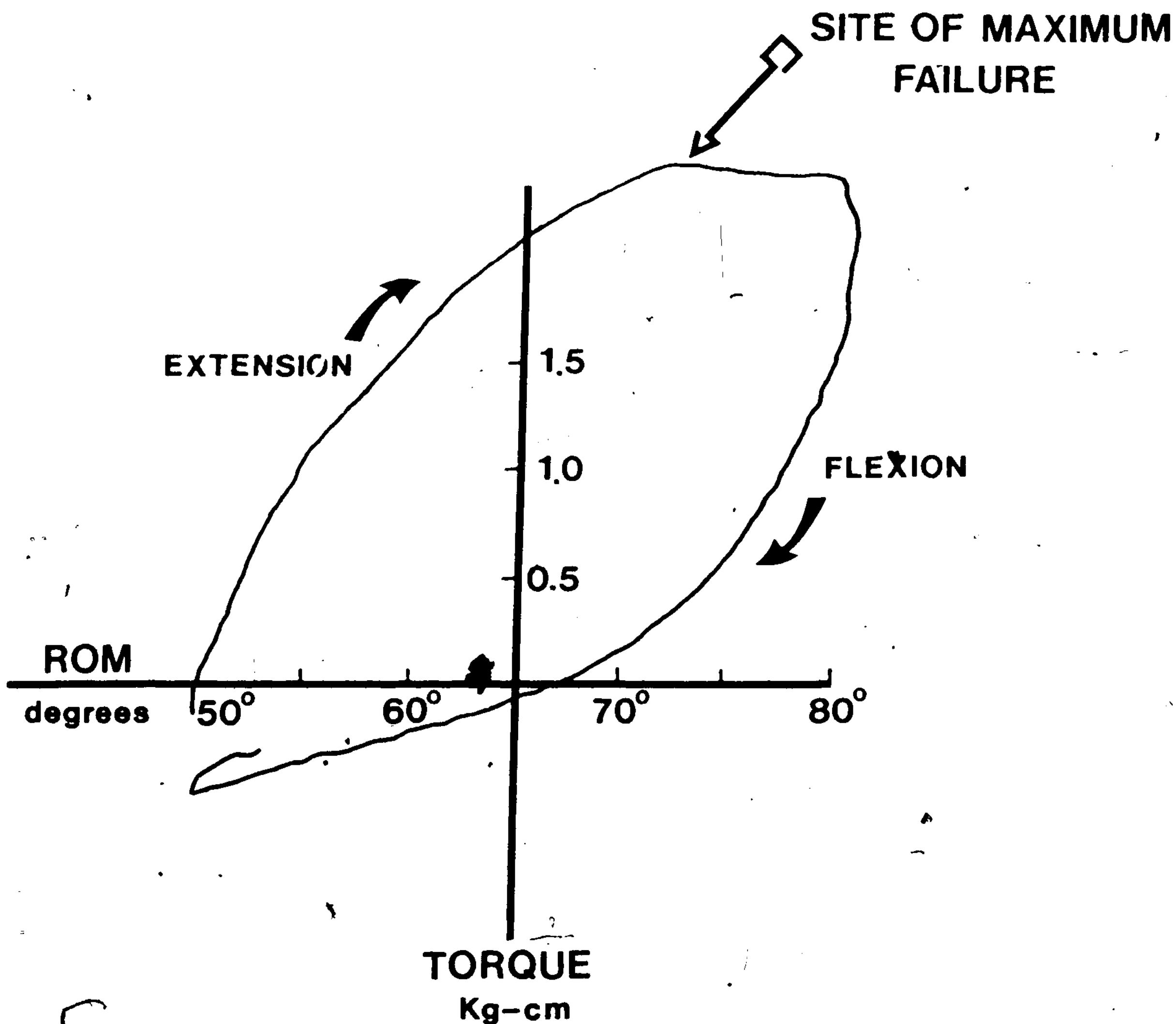


FIGURE IV-6 Torque-angular Displacement Curve for Contracture Rabbit's Knee 1st Cycle. The hysteresis loop resembles the curve seen earlier in Figure II-8, but with the graph inverted to clearly locate the knee extension portion as the upper curve. Evidence of ligamentous failure (site of maximum failure) is noted in the last 7 to 8 degrees where the loading curve levels off parallel to the horizontal axis. (Adapted from Woo et al.[254]).

Due to the omission of both fundamental operational definitions and possibly incomplete statistical analyses of data, comparisons between studies are at best difficult.

4.6 Discussion of Statistical Tables

Results and statistics are presented in the form of tables with subheadings:

a. means (standard deviations)

b. ANOVA summary, and when applicable (c, d, e)

c. Scheffe comparisons of groups

d. Scheffe comparisons of cycles

e. Tukey tests.

Results for the two-factor analysis of variance are in Tables 1 - 6, while results for the one-factor analysis of variance are in Tables 7 - 10. The results are related to each of the stated hypotheses.

H_1: Areas of hysteresis for control knees are essentially the same for each group and for each of the three cycles.

Results and statistics in Table IV-1(a,b,c) compare the areas of hysteresis of control knees for the three treatment groups over three cycles. There were no statistically significant differences among the three treatment groups ($p>0.05$). However, statistically significant differences were noted between cycles 1 and 2 ($p = 0.00$), and between cycles 1 and 5 ($p = 0.00$). The absence of statistically significant differences between treatment groups supports hypothesis H_1 and confirms similar findings from previous researchers.[6, 13, 254]

In analyzing bone-ligament-bone specimens (via load-deformation cycles to a predetermined peak displacement), previous studies have reported a decrease in area of hysteresis loop with repeated cycling. [45, 234, 236] Despite the decrease, no mention was made of statistically significant differences between

TABLE IV 1a

Means (Standard Deviations) for
Areas of Hysteresis (mm^2) of Control Knees for
Three Groups over Three Cycles

Group	1st Cycle	2nd Cycle	3rd Cycle
Group 1 [illegible]	[illegible]	[illegible]	[illegible]
Group 2 [illegible]	[illegible]	[illegible]	[illegible]
Group 3 [illegible]	[illegible]	[illegible]	[illegible]

[illegible]

TABLE IV 1b

ANOVA Summary of
Comparisons for Areas of Hysteresis (mm^2) of Control Knees
for Three Groups over Three Cycles

Source of Variation	Sum of Squares	D.F.	Mean Squares	F Ratio	Prob.
Between					
Treatment Groups (T)	[illegible]	[illegible]	[illegible]		
[illegible]	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]
Within Groups					
[illegible]	[illegible]	[illegible]	[illegible]		
Within					
Cycles (C)	[illegible]	[illegible]	[illegible]		
1st, 2nd, 3rd	[illegible]	[illegible]	[illegible]	[illegible]	0.000*
T x C					
Interaction	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]
Within					
Cycle Error	[illegible]	[illegible]	[illegible]		

*Statistically significant (p < 0.05)

TABLE IV 1c

Scheffe Comparisons of
Unweighted Main Effects of the Three Cycles

Comparisons of Cycles	Contrast	F Ratio	Probability
1st & 2nd	0.78780E+03	26.281	0.000*
1st & 3rd	0.93068E+03	36.678	0.000*
2nd & 3rd	0.14288E+03	0.864	0.[illegible]1

*Statistically significant (p < 0.05)

areas. However, it should be remarked that Woo et al.[254] in discussing hysteresis loops (from torque angular displacement curves), identified no difference in area of hysteresis between the 1st and 5th cycle (testing control rabbits' knees).

The presence of decreasing area with repeated cycling (viscoelastic behaviour)[234, 237] has been described as a "transient softening" of the ligamentous substance, yielding decreasing peak loads as "softening" continued.[8, 12, 249, 252] With applied strain and strain rate remaining constant, this softening with repeated cycling was present with slight decreases in the peak load.

It would appear that the present investigator's findings of statistically significant differences between 1st and 2nd cycles, also between 1st and 5th cycles, support this hypothesis of "transient softening" (Figure IV-4). The difference between the results of this study and the study by Woo et al.[254] could be possibly explained by the effect of preconditioning as done by Woo et al.[254]

H_2: Areas of hysteresis for treated knees vary from group to group and cycle to cycle. Regardless of cycle, the areas of the immobilised-only group (Group 1) are largest, with the areas of the exercised joint-mobilised group (Group 3) the smallest. The areas of the exercised group (Group 2) are slightly larger than those of Group 3. For all groups, the first cycle area is the largest, with each subsequent cycle area being much smaller.

Results and statistics in Table IV-2(a,b,c,d,e) compare the areas of hysteresis for treated knees (of the three treatment groups) over three cycles. A statistically significant difference (p = 0.046) occurred between Groups 2 and 3. Comparing the three cycles, there were statistically significant differences between both 1st and 2nd cycles (p = 0.00) and also between 1st and 5th cycles (p = 0.00). Comparisons of 1st cycle areas of hysteresis showed statistically significant differences between Groups 2 and 3 (p <0.05) and Groups 1 and 3 (p <0.05). The value of the means for Group 3 1st cycle areas of hysteresis was the smallest, while the value for Group 2 was the largest. Further significance

TABLE IV 2a

Means (Standard Deviations) for Areas of Hysteresis (mm^2) of Treated Knees for Three Groups over Three Cycles

Group	1st Cycle	2nd Cycle	5th Cycle
Group 1[7] (Immob only)	18,299.86 (7,053.83)	1,989.43 (603.17)	1,130.29 (324.93)
Group 2[6] (Ex)	20,401.66 (6,824.63)	3,628.83 (1,495.24)	2,539.67 (1,192.00)
Group 3[6] (Ex + Mob)	11,452.83 (3,188.60)	1,899.00 (726.11)	1,313.67 (580.43)

( ) Number in circle = number of subjects in group

TABLE IV 2b

ANOVA Summary of Comparisons for Areas of Hysteresis (mm^2) of Treated Knees for Three Groups over Three Cycles

Source of Variation	Sum of Squares	D.F.	Mean Squares	F Ratio	Prob.
Between Treatment Groups (T)	0.4440E+09	18	0.2467E+08		
1, 2, 3	0.1497E+09	2	0.7485E+08	3.974	0.040*
Within Groups					
1, 2, 3	0.3014E+09	16	0.1884E+08		
Within Cycles (C)	0.3320E+10	38	0.8737E+08		
1st, 2nd, 5th	0.2705E+10	2	0.1353E+10	103.585	0.000*
T x C Interaction	0.1455E+09	4	0.3638E+08	2.786	0.043*
Within Cycle Error	0.4179E+09	32	0.1306E+08		

*Statistically significant ($p < 0.05$)

TABLE IV 2c

Scheffe Comparisons of Unweighted Main Effects of the Three Cycles

Comparisons of Cycles	Contrast	F Ratio	Probability
1st & 2nd	0.14212E+05	73.474	0.000*
1st & 5th	0.15057E+05	82.466	0.000*
2nd & 5th	0.84455E+03	0.259	0.773

*Statistically significant ($p < 0.05$)

TABLE IV 2d

Scheffe Comparisons of
Unweighted Main Effects of the Three Treatment Groups

Comparisons of Groups	Contrast	F Ratio	Probability
1st & 2nd	0.17169E+04	0.758	0.485
2nd & 3rd	0.39682E+04	3.762	0.046*
1st & 3rd	0.22514E+04	1.304	0.299

*Statistically significant ($p < 0.05$)

TABLE IV 2e

Tukey Procedure for Comparisons of
1st Cycle Areas of Hysteresis of Three Groups

Group	J	df_w	q
2 3	3	16	5.05*
1 3	3	16	4.01*
2 1	3	16	1.23

*statistically significant ($p < 0.05$, $q > 3.65$)

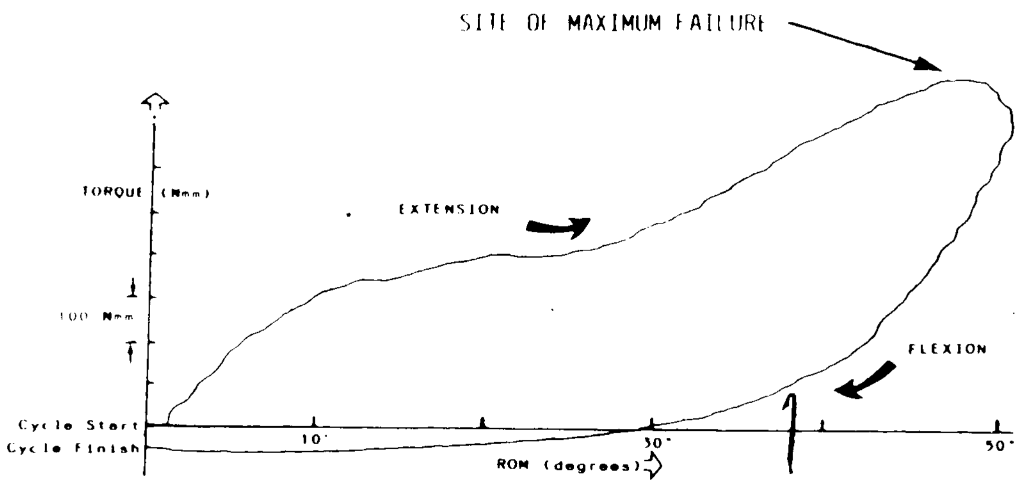


FIGURE IV-7 Torque-angular Displacement Curve (Rabbit #39 Ex Jt-Mob) 1st Cycle. The site of maximum failure occurs late in the knee extension portion of the curve. The slope of this curve increases slowly from the beginning to near the end of the range before levelling off in the last few degrees. There is evidence of failure throughout the total 50° ROM.

of these findings is summarized and discussed at the end of this section on the results of the statistical tables.

At the onset of the study, it was thought that dramatic differences in terms of 1st cycle of hysteresis would be evident between groups. Unexpected results were obtained, with obvious signs of capsular ligamentous failure. The most severe destruction appeared to occur in the 1st cycle loops of Group 1; whereas, in the same cycle, measurably less damage was sustained in Group 2. In Group 3 (1st cycle), appreciably less damage was sustained than in Group 1 (Figures IV-1, 2, 3, 7). The marked differences in areas of hysteresis between 1st and 2nd cycles, and between 1st and 5th cycles were also noted by Woo et al. [254]

It would appear from the 1st cycle graphs (Figures IV-1, 2) that Group 1 did undergo extensive tearing of the contracture soft tissues (extra and intra-articular structures). Surprisingly, this damage was not at all visually evident, an apparent incongruity that, nonetheless, is consistent with the findings of Kennedy et al.[120] Their findings indicated that it was possible at maximum failure for a ligamentous specimen to appear intact visually, despite considerable destruction and disorganization of collagenous fibrils (as revealed by the scanning electron microscope).

H_3 The hysteresis area ratio of 1st cycle to 2nd cycle (treated knees as compared to control knees) is greatest for Group 1 and smallest for Group 3. The area ratio of Group 2 is slightly greater than that of Group 3.

Results and statistics in Table IV-3(a,b) compare the hysteresis area ratio, 1st to 2nd cycle (treated and control knees), of the three groups. There was a statistically significant difference between treated and control knees (p = 0.00). There were no statistically significant differences among the three treatment groups.

TABLE IV 3a

Means (Standard Deviations) for Hysteresis Area Ratio of 1st to 2nd Cycle for Treated and Control Knees of Three Groups

Group	Treated Area Ratio	Normal Area Ratio
Group 1 (7) (Immob only)	9.79 (3.83)	1.52 (.26)
Group 2 (6) (Fx)	6.50 (2.94)	1.42 (.23)
Group 3 (6) (Fx Jt Mob)	7.29 (3.16)	1.36 (.18)

( ) Number in circle = number of subjects in group.

TABLE IV 3b

ANOVA Summary of Comparisons for Hysteresis Area Ratio of 1st to 2nd Cycle for Treated and Control Knees of Three Groups

Source of Variation	Sum of Squares	D.F.	Mean Squares	F Ratio	Prob.
Between Treatment Groups (T)	0.1388E+03	18			
1, 2, 3	0.2003E+02	2	10.015	1.360	0.285
Within Groups 1, 2, 3	0.1178E+03	16	7.363		
Within Area Ratios (AR)	0.5303E+03	19			
Treated, Control	0.3908E+03	1	390.766	58.108	0.000*
T — AR Interaction	0.1716E+02	2	8.580	1.276	0.306
Within Area Ratios Error	0.1076E+03	16	6.725		

*Statistically significant ($p < 0.05$)

A partial comparison can be made between the results of Woo et al,[254] Akeson et al,[11] and the present findings:

1. The previous researchers analyzed five cycles (50° - 80° ROM) followed by five more cycles (45° - 95° ROM).

2. For 50° to 80° ROM, the 1st cycle area of hysteresis was 10x greater for the control knee area, while by the 5th cycle it had reduced to 3 times.

3. For 45° to 95° ROM the immobilised group's 1st cycle was 6 times greater than the control knee area; while by the 5th cycle the immobilised group had also reduced to 3 times.[254]

Following one week remobilisation[11] it was found that for the 50° - 80° arc of motion, the area of hysteresis was 8 times greater than the control and for 45° - 95° ROM, the area was 5 times greater than normal. However, for these remobilisation results, it is not stated if there was only one cycle of motion at 50° - 80° ROM or five cycles as in earlier work. [254]

H4: Torque or 'stiffness' is minimal for control knees and essentially the same for each treatment group.

Stiffness proved minimal for control knees and was essentially the same for each group. Because of the similarities in curves between groups and between cycles, statistics were not calculated for each group of control knees. Figure IV-4 illustrates a typical torque-displacement curve for a control knee over the varying cycles (1st, 2nd, and 5th). This curve can be contrasted with that produced by the arthrograph alone (Figure III-8).

H5: End-range stiffness slopes (torque) for treated knees demonstrate greatest stiffness in the 1st cycle of all groups and are much lower, starting with the 2nd cycle of all groups. Torque is greatest in Group 1 and least in Group 3, with Group 2 torque slightly more than that of Group 3.

Results and statistics in Table IV-4 (a,b,c) compare end-range stiffness for treated knees of three groups over three cycles (2nd, 5th, and 7th cycles). No statistically significant differences appeared between groups; however, there were significant differences between cycles. These differences appeared between the 2nd and 5th cycles ($p = 0.025$) and also between the 2nd and 7th cycles ($p = 0.00$). The difference between the 5th and 7th cycles was almost statistically significant ($p = 0.09$).

Conclusions from these results (Figure III-11) may not be warranted, as it has been clearly demonstrated that ligamentous failure has occurred in all 1st cycle hysteresis loops, with dramatic differences in hysteresis area of the 1st cycle as compared to the 2nd. Failure is evident also in the obvious differences in shape of the loops. The upshot of this is a distinct possibility that cycles subsequent to the 1st cycle are drastically altered in both shape and area because of damage sustained in the 1st cycle.

Results and statistics in Table IV-5 (a,b,c) compare mid-range stiffness for the three treated knees of three groups over three cycles (2nd, 5th, and 7th cycles). No statistically significant differences emerged between groups, yet did appear between cycles: 2nd to 5th ($p = 0.00$), 2nd to 7th ($p = 0.00$), and 5th to 7th ($p = 0.036$) (Figure III-11).

Results and statistics in Table IV-6 (a,b) compare end-range and mid-range stiffness cycles for treated knees of the three groups over the 2nd cycle. There was a statistically significant difference between end-range stiffness results and mid-range stiffness results ($p = 0.001$).

End-range stiffness measurements (last 6° of motion) could not be made for first cycle loops, because nearly all hysteresis loops exhibited points of maximum failure before attaining end-range.

The absence of differences between stiffness measurements (mid-range and end-range) for the three groups over three cycles (2nd, 5th, and 7th) can be explained in terms of 1st cycle ligamentous destruction and the subsequent cycling of specimens after failure.

TABLE IV-4a

Means (Standard Deviations) for End-range Stiffness Cycles (mm) of Treated Knees of Three Groups over Three Cycles**

Group	2nd Cycle	5th Cycle	7th Cycle
Group 1(7) (Immob only)	28.71 (15.59)	27.43 (15.05)	26.14 (13.98)
Group 2(5) (Ex)	25.20 (5.91)	24.80 (5.85)	24.40 (6.09)
Group 3(6) (Ex Jt-Mob)	26.67 (7.07)	25.17 (6.77)	24.33 (6.45)

( ) Number in circle = number of subjects in group

**To convert end-range stiffness from millimeters to Nmm multiply the millimeter value by 7.88N.

TABLE IV-4b

ANOVA Summary of Comparisons for End-range Stiffness Cycles (mm) of Treated Knees for Three Groups over Three Cycles**

Source of Variation	Sum of Squares	D.F.	Mean Squares	F Ratio	Prob.
Between					
Treatment Groups (T)	0.6047E+04	17	355.706		
1, 2, 3	0.6720E+02	2	33.602	0.084	0.920
Within Groups					
1, 2, 3	0.5976E+04	15	398.388		
Within					
End Range Cycles (ERC)	0.7800E+02	36	2.167		
2nd, 5th, 7th	0.3208E+02	2	16.042	13.190	0.000*
T - ERC					
Interaction	0.5727E+01	4	1.432	1.177	0.341
Within					
ERC Error	0.3649E+02	30	1.216		

*Statistically significant ($p < 0.05$)

**To convert end-range stiffness from millimeters to Nmm, multiply the millimeter value by 7.88N.

TABLE IV-4c

Scheffe Comparisons of Unweighted Main Effects of the Three Cycles

Comparisons of Cycles	Contrast	F Ratio	Probability
2nd & 5th	0.10619E+01	4.172	0.025*
2nd & 7th	0.19016E+01	13.379	0.000*
5th & 7th	0.83968E+00	2.609	0.090*

*Statistically significant ($p < 0.05$)

TABLE IV-5a

Mean (Standard Deviations) for Mid range Stiffness Cycles (mm) of Treated Knees of Three Groups over Three Cycles**

Group	2nd Cycle	5th Cycle	7th Cycle
Group 1(7) (Immob only)	19.14 (11.58)	12.71 (8.75)	10.71 (7.20)
Group 2(5) (Ex)	22.60 (4.96)	16.40 (4.92)	15.20 (4.45)
Group 3(6) (Ex Jt Mob)	16.17 (5.58)	12.33 (5.65)	10.67 (5.47)

( ) Number in circle = number of subjects in group

**To convert mid-range stiffness from millimeters to Nmm, multiply the millimeter value by 7.88N.

TABLE IV-5b

ANOVA Summary of Comparisons for Mid range Stiffness Cycles (mm) of Treated Knees for Three Groups over Three Cycles**

Source of Variation	Sum of Squares	D.F.	Mean Squares	F Ratio	Prob.
Between					
Treatment Groups (T)	0.2865E+04	17	168.529		
1, 2, 3	0.2439E+03	2	121.946	0.692	0.516
Within Groups					
1, 2, 3	0.2643E+04	15	176.208		
Within					
Mid range Cycles (MRC)	0.6200E+03	36	17.222		
2nd, 5th, 7th	0.4904E+03	2	245.188	77.174	0.000*
T × MRC					
Interaction	0.1742E+02	4	4.354	1.371	0.268
Within					
MRC Error	0.9531E+02	30	3.177		

*Statistically significant ($p < 0.05$)

**To convert mid-range stiffness from millimeters to Nmm, multiply the millimeter value by 7.88 N.

TABLE IV-5c

Scheffe Comparisons of Unweighted Main Effects of the Three Cycles

Comparisons of Cycles	Contrasts	F Ratio	Probability
2nd & 5th	0.54873E+01	42.648	0.000*
2nd & 7th	0.71095E+01	71.592	0.000*
5th & 7th	0.16222E+01	3.727	0.036*

*Statistically significant ($p < 0.05$)

TABLE IV-6a

Means (Standard Deviations) for End-range and Mid-range Stiffness Cycles (mm) of Treated Knees of Three Groups over One Cycle**

Group	End-range Stiffness/2nd Cycle	Mid-range Stiffness/2nd Cycle
Group 1 (7) (Immob-only)	28.71 (15.59)	19.14 (11.58)
Group 2 (5) (Ex)	25.20 (5.91)	22.60 (4.96)
Group 3 (6) (Ex Jt-Mob)	26.67 (7.07)	16.17 (5.58)

○ Number in circle = number of subjects in group

**To convert end-range and mid-range stiffness from millimeters to Nmm, multiply the millimeter value by 7.88N.

TABLE IV-6b

ANOVA Summary of Comparisons for End-range and Mid-range Stiffness Cycles (mm) of Treated Knees for Three Groups over One Cycle**

Source of Variations	Sum of Squares	D.F.	Mean Squares	F Ratio	Prob.
Between					
Treatment Groups (T)	0.3042E+04	17	178.941		
1, 2, 3	0.4897E+02	2	24.483	0.123	0.885
Within Groups					
1, 2, 3	0.2992E+04	15	199.483		
Within					
ERC, MRC	0.1103E+04	18	61.278		
2nd Cycle	0.5044E+03	1	504.380	17.424	0.001*
T − (ERC, MRC)					
Interaction	0.1098E+03	2	54.899	1.896	0.184
Within					
(ERC, MRC) Error	0.4342E+03	15	28.948		

*Statistically significant ($p < 0.05$)

**To convert end-range and mid-range stiffness from millimeters to Nmm, multiply the millimeter value by 7.88N.

H_6: First cycle stiffness slopes (torque) of the treated knees are greatest for Group 1 and least for Group 3, with Group 2 slightly more than Group 3.

Results and statistics in Table IV-7 (a,b,c) compare 1st cycle stiffness slopes (torque) of treated knees for three groups. Statistically significant differences were in evidence between Groups 1 and 2 ($p = 0.049$) and also between Groups 1 and 3 ($p = 0.037$). No significant differences were seen between Groups 2 and 3.

To analyze the point of maximum failure further, 1st cycle vertical displacement and angular displacement to failure were examined more closely (Figure III-10).

Results and statistics in Table IV-8 (a,b) compare 1st cycle vertical displacement at point of linear load for three groups of treated knees. No statistically significant differences were found among these three groups.

Results and statistics in Table IV-9 (a,b,c) compare 1st cycle angular displacement at point of linear load for three groups of treated knees. Statistically significant differences were found between Groups 1 and 2 ($p = 0.016$) and also Groups 1 and 3 ($p = 0.015$). There were no statistically significant differences between the results from Groups 2 and 3.

The mean results of angular displacement at failure (degrees within the 50° total ROM) were:

20.59° for Group 1

38.40° for Group 2

38.60° for Group 3.

Group 1 failed on average at 41% of total range, while Groups 2 and 3 failed at 77% of total range — in other words, at less than half the total range and at approximately three-quarters the total ROM.

Results and statistics in Table IV-10 (a,b,c) compare displacement at failure, expressed as the ratio of vertical to angular displacement for 1st cycle treated knees of three groups.

TABLE IV-7a

**Means (Standard Deviations) for
1st Cycle Stiffness Slopes (Nmm) for
Treated Knees of Three Groups**

	(Immob-only) Group 1⑦	(Ex) Group 2⑥	(Ex Jt-Mob) Group 3⑥
1st Cycle Slopes	61.33 (25.43)	32.04 (12.42)	30.42 (9.87)

○ Number in circle = number of subjects in group.

TABLE IV-7b

**ANOVA Summary of
Comparisons for 1st Cycle Stiffness Slopes (Nmm) of
Treated Knees for Three Groups**

Source of Variations	Sum of Squares	D.F.	Mean Squares	F Ratio	Prob.
Between Treatment Groups (T) 1, 2, 3	0.4013E+04	2	0.2006E+04	5.32	0.017*
Within Cycle Error	0.6037E+04	16	0.3772E+03		
Total	0.1005E+05				

*Statistically significant ($p < 0.05$).

TABLE IV-7d

**Scheffe
Post-hoc Pairwise Contrasts of the Three Groups**

Group	Mean Difference	Standard Error	Lower	Upper	D.F.1	D.F.2	F	$\frac{F}{(J-1)}$	Prob.
1 - 2	-29.29	116.78	-58.43	-0.15	2	16	7.35	3.67	0.049*
2 - 3	1.61	125.76	-28.63	31.86	2	16	0.02	0.01	0.990
1 - 3	30.90	116.78	1.76	60.05	2	16	8.18	4.09	0.037*

*Statistically significant ($p < 0.05$)

TABLE IV-8a

Means (Standard Deviations)
for 1st Cycle Vertical Displacement (mm) at Point of**
Linear Load for Three Groups of Treated Knees

	(Immob. only) Group 1(7)	(Ex) Group 2(6)	(Ex-H Mob) Group 3(6)
Vertical Displacement	91.29 (32.[illegible])	118.33 (32.63)	81.00 (22.99)

( )Number in circle = number of subjects in group

**To convert vertical displacement from millimeters to Nmm, multiply the millimeter value by 7.88N.

TABLE IV-8b

ANOVA Summary of
Comparisons for 1st Cycle Vertical Displacement (mm) at**
Point of Linear Load for Three Groups of Treated Knees

Source of Variation	Sum of Squares	D.F.	Mean Squares	F	Prob
Between Treatment Groups (T) 1, 2, 3	0.3746E+04	2	1,872.80	1.75	0.205
Within Cycle Error	0.1708E+05	16	1,067.55		
Total	0.2083E+05				

*Statistically significant (p < 0.05)

**To convert vertical displacement from millimeters to Nmm, multiply the millimeter value by 7.88N.

TABLE IV-9a

Means (Standard Deviations) for 1st Cycle Angular Displacement (cm) at Point of Linear Load for Three Groups of Treated Knees**

Angular Displacement	(Immob only) Group 1(7)	(Ex) Group 2(6)	(Ex Jt Mob) Group 3(6)
cm	10.40 (4.34)	19.42 (3.14)	19.52 (5.73)
degrees	20.59 (8.59)	38.45 (6.22)	38.65 (11.35)

( ) Number in circle = number of subjects in group

**To convert angular displacement from centimeters to degrees, multiply centimeter value by 1.98

TABLE IV-9b

ANOVA Summary of Comparisons for 1st Cycle Angular Displacement (cm) at Point of Linear Load for Three Groups of Treated Knees**

Source of Variation	Sum of Squares	D.F.	Mean Squares	F Ratio	Prob.
Between Treatment Groups (I) 1, 2, 3	0.3635E+03	2	181.73	7.48	0.005*
Within Cycle Error	0.3887E+03	16	24.29		
Total	0.7522E+03				

*Statistically significant ($p < 0.05$)

**To convert angular displacement from centimeters to degrees, multiply the centimeter value by 1.98°.

TABLE IV-9d

Scheffe Post hoc Pairwise Contrasts of the Three Groups

Group	Mean Difference	Standard Error	Lower	Upper	DF1	DF2	F	F/(J-1)	Prob.
1 - 2	- 9.02	7.52	1.62	16.41	2	16	10.81	5.41	0.016*
2 - 3	0.10	8.10	-7.77	7.57	2	16	0.00	0.00	0.999
1 - 3	- 9.12	7.52	-16.51	-1.72	2	16	11.05	5.53	0.015*

*Statistically significant ($p < 0.05$)

TABLE IV 10a

Means (Standard Deviations) for
Ratio of Vertical to Angular Displacement at Point of Linear Load During the 1st Cycle for Three Groups of Treated Knees

	(Immob only) Group 1 (7)	(Ex) Group 2 (6)	(Ex Jt Mob) Group 3 (6)
Ratio Vertical to Angular Displacement	.982 (.325)	.639 (.233)	.434 (.060)

( ) Number in circle = number of subjects in group

TABLE IV 10b

ANOVA Summary of
Comparisons for Displacement at Failure as Expressed in a Ratio of Vertical to Angular Displacement for 1st Cycle Treated Knees of Three Groups

Source of Variation	Sum of Squares	D.F.	Mean Squares	F Ratio	Prob.
Between Treatment Groups (1) 1, 2, 3	0.1006E+01	2	0.50	7.46	0.005*
Within Ratio Error	0.1078E+01	16	0.07		
Total	0.2084E+01				

*Statistically significant ($p < 0.05$)

TABLE IV 10d

Scheffe
Post hoc Pairwise Contrasts of the Three Groups

Group	Mean Difference	Standard Error	Lower	Upper	DF1	DF2	F	$\frac{F}{(J-1)}$	Prob
1 2	0.34	0.02	0.73	0.05	2	16	5.65	2.83	0.089
2 3	0.21	0.02	-0.20	0.61	2	16	1.88	0.94	0.412
1 3	0.55	0.02	0.16	0.94	2	16	14.43	.22	0.006*

*Statistically significant ($p < 0.05$)

Earlier findings of angular displacement did produce statistically significant findings between Groups 1 and 2 and also between Groups 1 and 3. Vertical displacement findings were not statistically significant. However for ratio of vertical to angular displacement values statistically significant differences were found between Groups 1 and 3 ($p = 0.006$), with approaching significance between Groups 1 and 2 ($p = 0.089$).

Table IV-11 summarizes the relationship between vertical and angular displacement along with area size. To clarify these differences further, Figure IV-8 illustrates vertical displacement at failure, angular displacement at failure, 1st cycle slopes and, to a certain extent, area size under the linear loading cycle.

The significance of the 1st cycle hysteresis loops in terms of area and stiffness measurements is very clearly shown on the graph (Figure IV-8). Group 1 presents with a sharp increase in the slope in the linear region and fails earlier in the 50° range (at 20.59°), but with a greater 1st cycle area. Both Groups 2 and 3 fail at a similar point in the 50° range (at 38.4° and 38.6°). However, the vertical displacement of Group 2 is much higher, as shown by the long steep slope in the linear region. Due to this steeper slope, the underlying area of Group 2 is much larger than that area underlying the curve of Group 3.

Table IV-12 illustrates the hysteresis loop area sizes for each cycle in turn. The area of Group 1 for the 1st cycle is 7.5 times greater than the control area; this relationship by the 2nd cycle, has decreased to 1.25 times greater than the control area. For Group 2, the area of the 1st cycle is 6.7 times greater than the control, and this relationship has decreased to 1.7 times in the 2nd cycle. The 1st cycle area for Group 3 is 4.5 times greater than the control, but is the same area by the 2nd cycle.

These comparisons illustrate the dramatic differences between 1st and 2nd cycle areas for each individual group. Identifying the area difference between the 1st and 2nd cycles in Figure I-1, one can more clearly observe the destruction taking place in the 1st cycle. By the 5th cycle there is very little difference between treated and control knees.

TABLE IV-11

Comparisons of Vertical Displacement, Angular Displacement and 1st Cycle Area Size for Three Groups

	Vertical Displacement (at failure)	Angular Displacement (at failure)	Area Size For Entire 50° Cycle	Vertical vs. Angular Displacement
Group 1	186.69 Nmm	20.59°	7.5x control	9.8
Group 2	234.29 Nmm	38.45°	6.7x control	6.4
Group 3	166.32 Nmm	38.65°	4.5x control	4.3

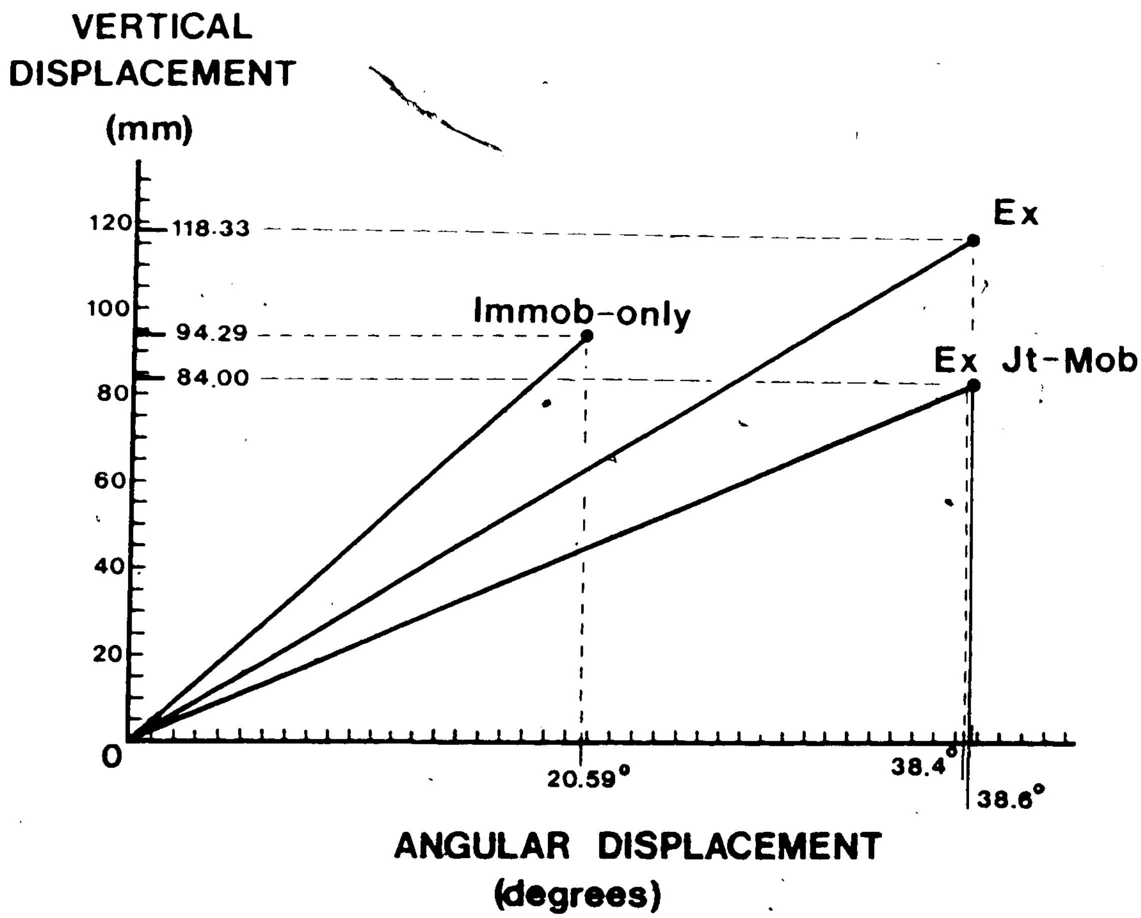


FIGURE IV-8, Comparisons of Vertical and Angular Displacements of Three Treatment Groups. The significance of the 1st cycle hysteresis loops in terms of angular displacement and vertical displacement is discussed in the text. To convert vertical displacement from millimeters to Nmm the millimeter value is multiplied by 7.88 N.

TABLE IV-12

Descriptions of Hysteresis Loop Areas

Comparing 1st Treated Knee Cycle to 1st Control Knee Cycle
(comparing group hysteresis means)

Group 1	(Immob-only)	Rx area = 7.5x control area
Group 2	(Ex)	Rx area = 6.7x control area
Group 3	(Ex Jt-Mob)	Rx area = 4.5x control area

Same Repeated for 2nd Cycle

Group 1	(Immob-only)	Rx area = 1.3x control area
Group 2	(Ex)	Rx area = 1.7x control area
Group 3	(Ex Jt-Mob)	Rx area = control area

Same Repeated for 5th Cycle

Group 1	(Immob-only)	Rx area = .71x control area
Group 2	(Ex)	Rx area = 1.3x control area
Group 3	(Ex Jt-Mob)	Rx area = .77x control area

Comparing Means of a Total Sum of Individual Ratios
1st to 2nd Cycle for Treated Knees

Group 1	(Immob-only)	1st to 2nd cycle ratio = 9.8
Group 2	(Ex)	1st to 2nd cycle ratio = 6.5
Group 3	(Ex Jt-Mob)	1st to 2nd cycle ratio = 7.3

To summarize, statistically significant results for 1st cycle findings were:

i) areas of hysteresis for treated knees

between Groups 2 and 3 ($p < 0.05$)

between Groups 1 and 3 ($p < 0.05$)

ii) first cycle stiffness slopes

between Groups 1 and 2 ($p = 0.049$)

between Groups 1 and 3 ($p = 0.037$)

iii) angular displacement

between Groups 1 and 2 ($p = 0.016$)

between Groups 1 and 3 ($p = 0.015$)

iv) ratio of vertical to angular displacements (rise/run)

between Groups 1 and 3 ($p = 0.006$)

and almost statistically significant between Groups 1 and 2 ($p = 0.089$).

The results for Group 3 for 1st cycle findings as compared to Groups 1 and 2 were:

i) statistically significant smallest area of hysteresis

ii) differences in means, though not statistically significant, with:

- least vertical displacement for Group 3
- least stiffness for Group 3
- smallest ratio vertical to angular displacement for Group 3

iii) almost identical angular displacement, with Group 3 slightly further into the range.

The 1st cycle area of hysteresis was largest for the exercised group and smallest for Group 3. The failure of Group 1 fairly early in the range (at less than half the full range) as compared to Groups 2 and 3 (at about three-quarters the full range) resulted in a smaller area of hysteresis than originally anticipated. The hypothesis (H_2) did not take into consideration the possibility of ligamentous failure during the 1st cycle.

Because of early failure of immobilised-only specimens, the first cycle slope measurement (linear region) becomes even more critical - mid-range and end-range stiffness measurements are no longer applicable. During the first cycling motion, there was most likely tearing of i) fibrofatty tissue adherent to the articular tissue, ii) anomalous crosslinks, iii) synovial fold adhesions, and iv) collagenous fibre intercept point binding sites.

After only one week of remobilisation, or one week of remobilisation in conjunction with specific joint-mobilisation treatments, significant differences were apparent between the three groups.

It would appear that less tissue damage occurred during cycling of the knees of Groups 2 and 3. As a result of their treatments, the tissues were more pliable for Groups 2 and 3.

As noted previously, motion is vital for the synthesis of proteoglycans and collagen. It had been previously observed that the water and hexosamine (sugar residue of GAGS) levels had increased dramatically after one week of remobilisation.[11] With a partial recovery of water and hexosamine concentrations, motion should enable the process of lubrication and maintenance of critical distance between existing collagenous fibres to begin more effectively. As compared to the other groups, the immobilised-only group demonstrated the steepest slope in the 1st cycle, due to the relative absence of motion prior to testing.

During motion (exercise and joint-mobilisation), extraneous intermolecular and intramolecular crosslinks are broken down. Fibrous adhesion sites at the articular cartilage, fibrous proliferation within the joint space, adherent synovial folds, and overlapping collagenous fibres are stressed and stretched. Joint-mobilisation functioning more locally should enhance these changes even more specifically than the exercised group.

During joint-mobilisation, there should also be a direct effect on the collagenous sheaths surrounding the ligament (endotenon, epitenon, paratenon). These sheaths run perpendicular to the long axis of the ligament. No mention has been made of the effect of immobilisation on these sheaths.

However, one could postulate that the normal 'binding'[60] function of these sheaths would become greatly exaggerated.

The fibre meshwork of joint capsules is loose enough to allow the required flexibility for movement. When collagenous fibres are laid down or replaced (synthesis or degradation), the length and mobility of ~~these~~ fibres between attachments is dependent upon motion of the fibres during the period of formation.[127] If there is no motion, the fibres will tend to shorten and form dense rather than loose meshworks.

During immobilisation, there is an increase in rate of collagen synthesis, collagen degradation, and rate of crosslink formation.[4, 5, 10] The relatively immature collagen and crosslinks should respond well to motion. As the collagen matures during remobilisation, anomalous crosslinks should break down and the remaining crosslinks organize appropriately along lines of applied stress.

It has been postulated that joint-mobilisation will specifically restore the loss in anterior tibial gliding in relation to the femur,[117] provide a specific lubrication effect for the joint surfaces,[124] and locally stretch the joint capsule and periarticular joint structures.[172] The additional benefits of joint-mobilisation were decidedly apparent in 1st cycle results.

A more specific description of how joint-mobilisation increases available joint range and subsequently decreases joint stiffness may include:

i) stretching of the entire multi-directional meshwork of the joint capsule through traction;

ii) anterior and posterior stretching of the joint capsule during anterior tibial gliding (increasing mobility of collagenous fibres in the direction required for knee extension);

iii) stretching of: intra-articular fibrous tissue, ligamentous sheaths, sites of abnormal crosslinks, and scar tissue (developing between layers of capsular meshwork); and

iv) lubrication of the joint, meniscii, and ligaments.

4.7 Clinical Relevance

1. The lack of accurate measuring tools for physiotherapists to quantify joint stiffness is very evident. With regard to subjective measurements, Rhind et al[181] found that patients were incapable of distinguishing between joint stiffness and pain. As well, these same patients were unable to define the symptoms of stiffness in a consistent manner.

As available objective measurements, the Stoddard-Paris joint motion classification and Maitland's movement diagrams are a means of semi-quantitatively measuring joint stiffness.[82] However, both of these measurements are based on the application of well-developed manual skills.

In this particular study, both groups 2 and 3 failed at a similar point in the 50° range (at 38.4° and 38.6°). Despite the similarity of the total range of motion, the quality of the stiffness present throughout the range was decidedly different. The use of Maitland's movement diagrams would adequately record these differences. However, not everyone will develop excellent skills in order to appreciate the quality of this joint movement.

The need for an arthrograph suitable for clinical use becomes extremely apparent. With a more accurate documentation of initial joint stiffness, physicians and physiotherapists alike would be able to note any improvement in the quality of movement as a result of the applied treatment.

Arthrographs have been developed for clinical use as well as for scientific usage. However, these arthrographs have been developed only for the knee[90, 210, 215] and metacarpophalangeal (MCP) joints.[107, 225, 259] More recently, an even more portable microprocessor-controlled arthrograph has been developed for the MCP joint.[98] Unfortunately, these arthrographs are joint specific and only for these two joints.

2. This study confirms the beneficial effects of joint-mobilisation and exercise to decrease joint stiffness throughout the range (Figure IV-8).

With repeated graded joint-mobilisation movements of either traction or gliding, a physiotherapist senses during the treatment a gradual increase in the range of motion and a decrease in the joint resistance felt. In other words, there is probably increasing elongation of the joint capsule with decreasing amounts of required force.

Each repeated grade movement (Grades 3 and 4) could be likened to repeated hysteresis loops within the linear region (Figure II-7). As these loading and unloading cycles are repeated, the strain increases with decreasing stress levels. As well, the area of hysteresis becomes smaller and smaller.[237]

How far into this linear region graded traction or gliding movements occur is unknown. Paris[172] reasons that if joint-mobilisations and joint-manipulations remain within the elastic region, the connective tissue will return to its original resting length once the stress is removed. Furthermore, to gain ROM, the joint must be stretched into the plastic region, for a permanent deformation or lengthening has occurred on unloading. Viidik[237] stated that with repeated cyclic tests within the linear region, some of the change in length is reversible or recoverable (elastic effect), while the plastic component is not recoverable.

If permanent deformation results from joint-mobilisation, how long will this permanent elongation effect remain? If a joint is not used normally with its newly-acquired ROM, the joint will once again suffer from immobilisation effects. In other words, the patient must maintain new movement by means of home exercises, specific joint mobility exercises, as well as stretching exercises to maintain and increase the extensibility of the surrounding soft tissues.[29, 62, 127, 128, 135, 189]

Farfan[71] stressed the importance of strengthening exercises to help maintain the normal functioning of this newly-acquired ROM. It was much easier to prevent muscle and joint tightness and stiffness "by frequently repeated activity" than to correct it once it had developed.[127]

During a period of remobilisation, tendons and ligaments regain their ultimate strength at an extremely slow rate (Figure IV-9). Not only do tendons and ligaments take an extremely long time to recover, but there are also regions of periosteal absorption at the bone-ligament attachment points.[3, 250] It is unknown how long it would take for the periosteum to return to normal.

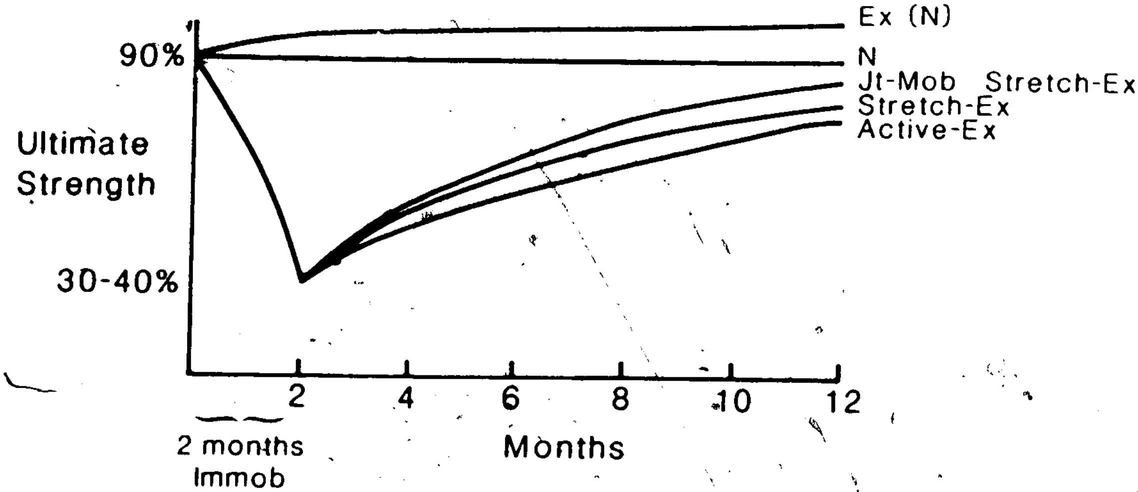


FIGURE IV-9 Ultimate Strength and Time Group Comparisons.

Following two months of immobilisation the conjectural recovery of treated ligamentous tissue improves the least for the active exercise (Active-Ex) group, greater for the muscle stretching with active exercise (Stretch-Ex) group and greatest for the active exercise with muscle stretching and joint-mobilisation (Ex' Jt-Mob Stretch Ex) group. Despite the intense treatment for these groups, even at the end of one year, the recovery has not reached the normal level and definitely not to the level achieved in the normal exercised group during the same time period. (Adapted and modified from Akeson et al.[8]).

"Prescribed exercises which increase the forces being transmitted to ligaments, tendons, and bones will maintain and generally increase the strength and functional capacity of these structures."[220] With our awareness of the deleterious effects of joint-immobilisation, it must be stressed to the patient to maintain an exercise level to increase joint mobility and to stretch surrounding soft tissues (muscles, fascia, and skin) in order to keep these tissues stretched as well as lubricated at the collagenous level.[76]

3. Within the limitations of this study, conclusions clearly indicate the beneficial effects of joint-mobilisation. This confirmation of beneficial effects on animal subjects needs to be reconfirmed on human subjects. Use of a rabbit model allows a consistency within the many variables present. The same control of the many variables would be next to impossible with human subjects. Furthermore, tests on the contribution of stiffness from just the joint complex (removing muscle, fascia, and skin) could not be done in the human.

4. The skills of physiotherapists, orthopaedic surgeons, mechanical engineers, rheumatologists, and patients, to mention a few, are needed in the development of an arthrograph. To further the studies on the effects of joint-mobilisation, the physiotherapist must work as a team with members of other professions. "As with many other areas in orthopaedics, it will be only through the benefits of applied research that the efficacy of clinical practice can be maximized."[76]

V. CONCLUSIONS AND RECOMMENDATIONS

5.1 Conclusions

a. Statistical and Observational Conclusions

After only one week of remobilisation, either by itself or in conjunction with joint mobilisation, differences became apparent between treatment groups. Benefits of joint mobilisation were particularly evident in the 1st cycle. In this cycle Group 3, as compared to Groups 1 or 2, had the smallest area of hysteresis (statistically significant between Groups 1 and 3, and Groups 2 and 3), least stiffness in the linear region (only statistically significant between Groups 1 and 3, and Groups 1 and 2), least vertical displacement at failure (no statistically significant differences), and the smallest ratio of vertical to angular displacement at failure (only statistically significant between Groups 1 and 3) (Figure IV-8).

Both Groups 2 and 3 failed at very nearly the same point in the range (38.4° and 38.6°, respectively); however, at this point, the joint stiffness for Group 3 was less than for Group 2 (with no statistically significant differences). This difference in joint stiffness was shown, as illustrated in Figure IV-8, with a higher vertical displacement at failure for Group 2 and likewise steeper slope (however no statistically significant differences were found). In other words, both groups had nearly identical ROM (motion), yet the stiffness of the joint (quality of motion) was substantially different.

At present, it is unknown when, or if, complete recovery of tendon or ligament strength can occur after extended immobilisation. It can be recalled that on the load-deformation curve for a bone-ligament-bone specimen (as modified from Noyes et al[166]) (Figure II-11), the group of monkeys remobilised (free active exercise) for 12 months, presented with a load-elongation curve that had not yet reached a 'normal' (control) level. Thus, despite free active exercise for 12 months, 'normal' levels of tendon and ligament strength had still not been reached.

As a result of the findings of the present study, the investigator speculates that a group allowed free active exercise — and receiving specific joint-mobilisation — would have achieved strength levels notably closer to the 'normal' curves than did the exercised only group. An even higher level of tendon or ligament strength might have been achieved. Moreover, the investigator is of the opinion that, with joint mobilisation, strength gains could have occurred at a faster rate and the level of strength ultimately attainable would have been significantly higher.

b. Projected Conclusions

Exercised normal ligamentous tissue, compared to *unexercised normal* ligamentous tissue, demonstrates greater stiffness, higher maximum load at failure, and greater maximum energy at failure (Figure II-11). Furthermore, Figure IV-9 outlines the recovery period for ligamentous tissue, following two months of immobilisation, and allows strength-comparisons with normal ligaments. There is a rapid loss of ligament strength with immobilisation, and yet an extremely gradual and long recovery of this strength occurs over a period of 12 months or more.[8, 166] *Exercised-normal* ligamentous tissue - EX (N) - is depicted at 100 per cent strength by one year's end; whereas *unexercised-normal* tissue (N) is depicted at 90 per cent in the same time.

There is a progressive increase in post-immobilisation ligament strength. This progression varies from:

i) least strong post-immobilisation group: active exercise only (Active-Ex), to

ii) stronger post-immobilisation group: active exercise + muscle stretching (Stretch-Ex), to

iii) the strongest post-immobilisation group: active exercise + muscle stretching + joint-mobilisation (Jt-Mob Stretch Ex) (Figure IV-9).

5.2 Recommendations

1. Cycling Range of Motion

a) The chosen ROM was unsuitable for this particular study. The immobilised group failed at 20.6°. The ideal ROM for testing joint stiffness would be well below the region of failure, preferably within the linear region.

b) As part of a further pilot study, small repeating loading-unloading cycles further and further into the range would isolate the desired range of motion. This particular approach has been described by Unsworth et al[225] for testing of the human MCP joints. Despite the preconditioning effects of these repeating cycles, these effects are desirable to cycling through too large a ROM.

c) Use of high speed motion pictures[45] or use of microscopy [120] could further document damage taking place during the cycling ROM.

2. Changes in Arthrograph

The ideal machine would not force the test specimen to follow the prescribed machine displacement, but would allow flexion and extension of the specimen to occur naturally. For more accurate results, a future arthrograph for testing the rabbit's knee should eliminate unnecessary forces from friction and gravity caused by shims, counterbalance, fixed machine axis point, and four fixation sites on the tibia and femur. The machine as it was did produce some undesirable forces and torsion on the knee joint.

The purpose of the teflon tubes was to allow physiological axial rotation or changing centre of rotation to occur without inducing unnecessary forces on the specimen. These tubes had a better "give" to them than the initial design with fixed pins into the bone. As the tubes accommodated to the ever-changing knee axis points, the machine axis point remained fixed. It could not be ascertained that friction in the teflon tubes was entirely eliminated.

As well, the onset of failure of the specimen could have been hastened by the initial knee position: i) tibia already rotated since the start of the immobilisation period, and ii) choosing of shims as well as the positioning of the limb within the tubes.

3. A tremendous amount of work has been done on testing biomechanical properties of tendons and ligaments in isolation, with much less emphasis on the joint capsule. Because testing by means of arthrograph involves the entire joint complex, the findings from these tests are more clinically

significant to the practitioner concerned about joint stiffness. Further research needs to be directed into the development of arthrographs both for clinical and laboratory research.

In summary, an improved experiment would begin with a small displacement cycle and would monotonically increase in magnitude. In this way, the minimum cycle causing damage could be identified, and the stiffness of the joint could be determined without damaging the specimen. As well, an improved testing apparatus would permit physiological testing. The specimen would not be forced to follow the trajectory of the machine and would not include friction from the machine components in its results.

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Personal Communication

1. Budney D: 1985
2. Reid DC: 1987
3. Secord DC: 1984
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APPENDIX A

RECORDING SUBJECTIVE MEASUREMENTS OF JOINT STIFFNESS

VISUAL ANALOGUE

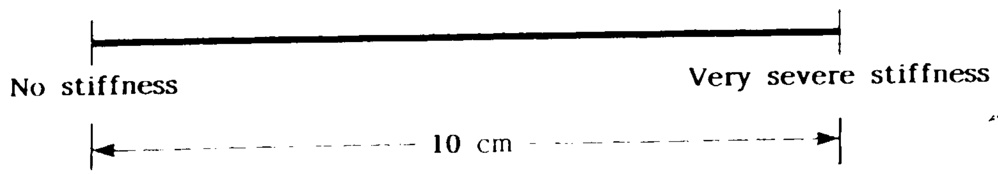

FIGURE A-1

The above solid, horizontal line represents a continuum of joint stiffness in terms of a uni-dimensional, ordinal scale with no stiffness at one end and very severe stiffness at the other end. The length of the line, or distance from 0 cm (no stiffness) corresponds to the amount of stiffness subjectively experienced. Accordingly, a subject marks the line and thus indicates their subjective perception of their joint stiffness.[42, 96, 181]

■ 5-POINT VERBAL SCALE

- ☐ No stiffness
- ☐ Mild stiffness
- ☐ Moderate stiffness
- ☐ Severe stiffness
- ☐ Very severe stiffness

FIGURE A-2

The above scale is an example of a 5-point verbal scale. It is a descriptive unidimensional, ordinal scale for recording joint stiffness where stiffness ranges in order of increasing severity, from no stiffness to very severe stiffness.[42, 96, 181]

■ NUMERICAL RATING SCALE

☐ 0 No stiffness

☐ 1

☐ 2

☐ 3

☐ 4

☐ 5

☐ 6

☐ 7

☐ 8

☐ 9

☐ 10 Very severe stiffness

FIGURE A-3

The above scale records joint stiffness as a unidimensional ordinal scale (with equal appearing intervals), where the number **0** indicates no stiffness and the number **10** indicates very severe stiffness. The number chosen by the patient corresponds to their subjective perception of their joint stiffness.[88, 96, 181]

■ GUTTMAN SCALE

- ☐ 1 Walking on a level surface
- ☐ 2 Walking on a slightly inclined, flat surface
- ☐ 3 Sitting onto a high stool
- ☐ 4 Ascending stairs
- ☐ 5 Putting on socks or nylons
- ☐ 6 Sitting crosslegged

FIGURE A-4

The above scale illustrates a Guttman scaling technique for knee ROM in which functional activities are sequenced in terms of increasing required ROM. The result is a unidimensional, cumulative scale which records joint stiffness. In terms of the scale, inability to perform one activity implies that it would be impossible to perform another activity further up on the scale; however, no difficulty should be experienced with performing an activity further down the scale.[21, 22, 23, 44] This hypothetical scale is untested for validity or reliability.

APPENDIX B

ANIMAL CARE COMMITTEE CONSENT

S.M.R.I. ANIMAL UNIT COMMITTEE RESEARCH/TEACHING PROJECT PROPOSAL

№ 0194

New Protocol [X]
Renewal [ ]
Modification [ ]

Previous Protocol No.

Date May 25 19 84

Senior Investigator or Course Director Dr. David Magee, Dr. David Secord — Phone No 5983, 3577

Department Physical Therapy, S.M.R. Animal Centre

Project Title The Effectiveness of Joint-mobilisation and Exercise on the Biomechanical Properties of the Immobilised Rabbit's Knee

Emergency Phone(s) 435-8868

Investigator(s) carrying out research Barbara E. Robinson — Phone No 5983, 435-8868

Department Physical Therapy

Please Check One: Research [X] Teaching [ ] Research and Teaching [ ]

Supporting Agency none at present

ANIMAL SPECIES	Approx. Number 35	Location of Experiment	Type of Experiment		Expected Pain Level		Anaesthetics and/or Analgesics to be used
New Zealand white rabbits	Per Month 35	Building Ellerslie	Surgical	X	Nil		anaesthetized with halothane
	2 yr. total	Room No.	Non-surg.		Low	X	
			Acute		Mod		
			Survival		High		

Methods of Euthanasia to be employed: intravenous overdose of pentobarbital sodium

Anaesthetic overdose	X
Cervical dislocation	
Decapitation	
Gas, Ether, CO_2	
Other	

Outlines of procedures and techniques to be used:
(Additional pages may be used if required)

OBJECTIVES: To compare the torque-angular displacement curves of a flexion-extension cycle for an immobilised rabbit's knee: a) immediately following a period of immobilisation, b) following a period of remobilisation of free active exercise and c) following a period of remobilisation with the addition of specific physiotherapeutic joint-mobilisation.

METHOD: Thirty-five male New Zealand white rabbits, each weighing approximately 3 kg (± 500 g), will be used as experimental animals. A 2.4 mm diameter stainless steel Steinmann pin will be inserted through the middle of the left tibia from anterior to posterior. The pin, remaining within the skin fold of the leg, will then be inserted through the middle of the proximal femur. An X-ray will verify the position of the pin.

The right knee will act as a control. Thirty of the rabbits will be maintained in this position of fixation for nine weeks. The remaining five rabbits will be immobilised in this position for seven weeks.

The 35 rabbits will be divided into one pilot study of five and into three groups of 10 each. During the time of immobilisation, the rabbits will be allowed to move around as they are able, in individual pens. The left ear of each rabbit will be tattooed with a number to avoid a mix-up between groups.

(continued next page)

Gold — Include with Grant Application
Blue — Retained by Director, SMRI Animal Unit
White — Investigator's copy

All animals used in this research project will be cared for in accordance with the recommendations of the Canadian Council on Animal Care, the requirements under Section 50 of the Universities Act and the regulations of the University Animal Welfare Policy Committee, the University of Alberta

Barbara E. Robinson
Principal Investigator

D. Secord May 28/
Director, SMRI ANIMAL UNIT — Date

Chairman, SMRI ANIMAL UNIT COMMITTEE — Date

After 9 weeks, the first group of ten rabbits will be sacrificed. A repeat X-ray just before sacrifice will ensure the maintenance of the fixation position during the period of immobilisation. The lower limbs will be disarticulated at the hip joint and all the skin of the hind limb removed. The muscles will be divided in a circumferential manner at approximately 4 cm above and 3 cm below the knee joint line. This circumferential incision will serve to eliminate the muscle and skin factors involved in the joint contracture created by the immobilisation.

The measuring instrument, a modified Wright-Johns-Goddard arthrograph, will measure the amount of torque required to extend and to flex the rabbit's knee through varying ranges of motion. A Hewlett-Packard Model 7041A X-Y recorder will yield the torque-angular displacement curve for this extension-flexion cycle.

Both the contracture and the control knee for each rabbit from group 1 will be tested on the arthrograph.

For both the second and third groups of 10 rabbits, the immobilisation fixation will be removed at the end of nine weeks. The rabbits will be allowed to run free in their individual pens for an additional three weeks. In addition, the third group of rabbits will receive specific joint-mobilisation treatments to the contracture knee three times per week. Both groups of rabbits will be sacrificed at the end of three weeks. The testing procedure as described for group 1 will be repeated for both the contracture and control knee for each rabbit.

For the pilot group of rabbits, the immobilisation fixation will be removed after seven weeks. After two weeks of free exercise and joint-mobilisation treatments three times per week the rabbits will be sacrificed and the biomechanical testing performed.

FEASIBILITY:

The pilot group of five rabbits will help to familiarize the investigator with the procedures involved and will identify future potential problems.

The Hewlett-Packard Model 7041A X-Y recorder and arthrograph can be obtained on loan from the physical therapy department.

The S.M.R.I. Animal Unit at Ellerslie is well equipped with the required pens and an isolated area far removed from other animals. Restraining jackets will be provided by the unit for the treatment sessions. The investigator will perform all the testing and treatment procedures. Surgical and radiological procedures will be performed with the assistance of a surgical technician and Dr. Secord.

APPENDIX C

MACHINE CALIBRATION

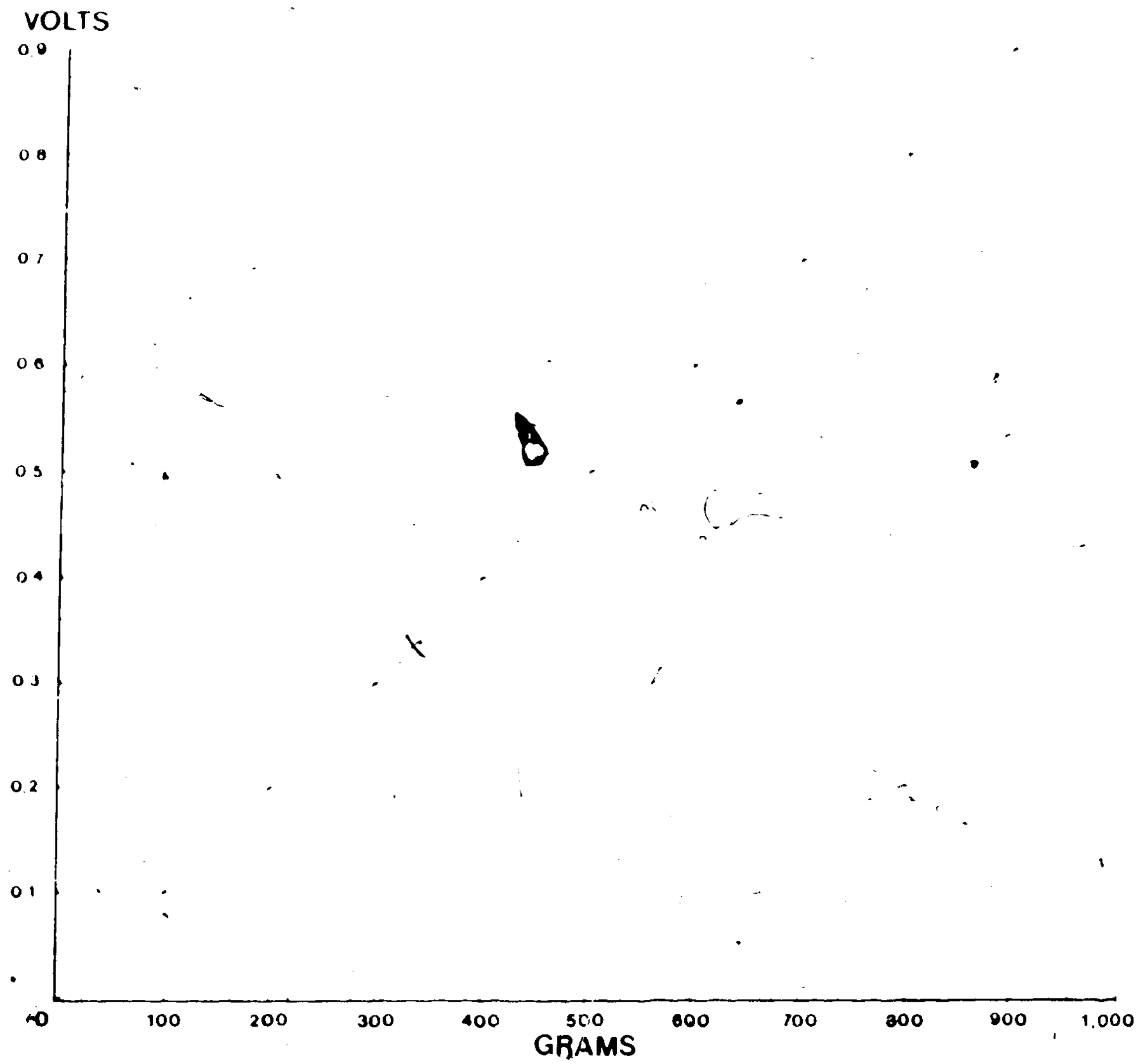

FIGURE C-1

Load Cell Calibration shows a linear relationship between voltage output and grams of force.

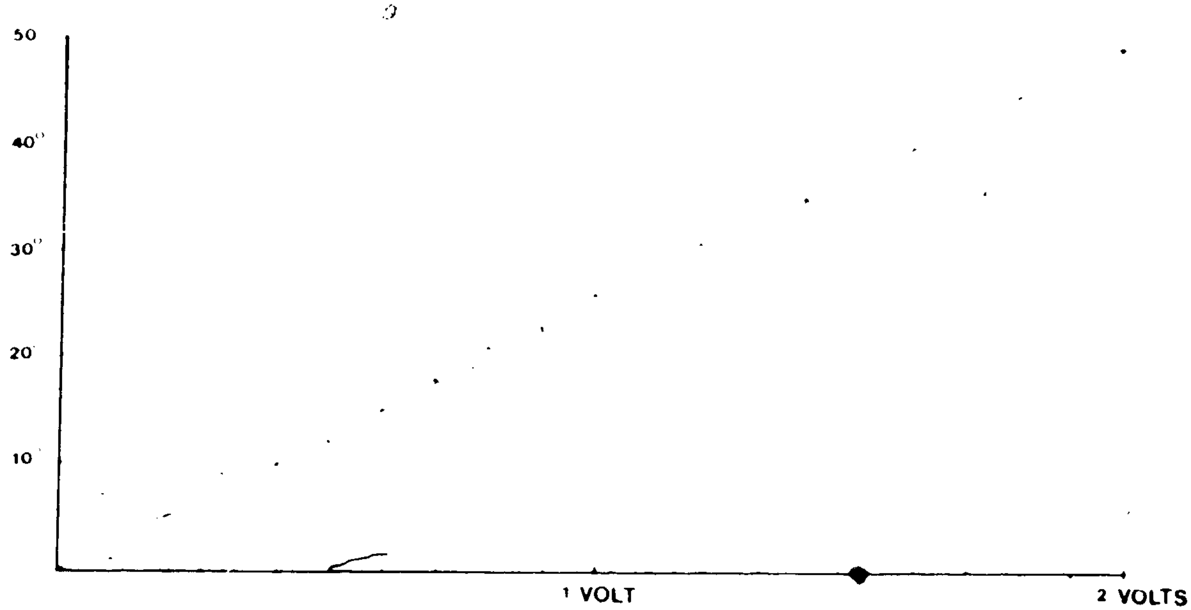

FIGURE C-2

Potentiometer Cailbration shows a linear relationship between voltage output and ROM in degrees.

APPENDIX D

IMMOBILISATION SURGICAL PROCEDURE

Immobilisation Surgical Procedure

The rabbit was first anaesthesized through a face mask, utilizing halothane with 3.5 per cent oxygen at a flow rate of 3 liters/minute. The entire left lower extremity was then shaved from groin to ankle. Following this, the shaved leg was prepared with soap and water wash and then sterilized by painting with a skin antiseptic, Betadine solution (U.S.P. topical solution 1 per cent iodine - 10 per cent povidone). All surgical instruments, gowns, and gloves had been previously sterilized in the autoclave.

The rabbit (except for its hind limb) was draped with two small sterilized drapes. One sterilized lap sponge enclosed the left foot. Both the lap sponge and two small drapes were held in place by seven Backhaus towel clips. Throughout surgery, the technician stabilized the left hind limb in a position of full knee flexion.

Two incisions were made. The first incision, just lateral to the tibial tuberosity, separated the tibialis anterior muscle from the tibia. The periosteum was separated medially, laterally, and a little inferiorly to the tibial tuberosity.

A second incision separated the lateral aspect of the quadriceps from the tensor fascia latae. A gentle opening of the Mayo scissors further separated the quadriceps from the tensor fascia latae, just enough to allow a finger under the belly of the quadriceps to separate the middle-proximal femoral attachment of the quadriceps from the bone. Bleeding was minimal.

The periosteum and tibialis anterior were retracted from the tibia, and the quadriceps plus tensor fascia latae were retracted from the anterior femur.

A 2.4 mm diameter stainless steel Steinmann pin, with truncated point, functioned as a drill bit for a hand bone drill and was inserted in this manner just below the tibial tuberosity (extending from anterior to posterior aspect of the tibia). After the pin pierced the tibia, careful drilling directed the pin through the mid-posterior femur.

To allow slightly easier access to the anterior aspect of the femur underneath the quadriceps, a ribben retractor was used to hold the soft tissues of the thigh and leg apart. Then, a hexagonal nut was threaded onto the tip of the Steinmann pin that pierced the anterior femur (and extended beyond by about 5 mm) and emerged under the quadriceps muscle. Another hexagonal nut was affixed onto the other end of the pin, located at the anterior aspect of the tibia. Then the anterior tibial nut was tightened slightly to bring the knee into complete flexion, while at the same time the position of the anterior femoral nut was maintained. The excess length of the pin was cut off anterior to the tibia (using wire-cutting pliers).

Using a 3-0 Dexon on a taper cutting needle, the various layers of tissue were stitched together, one layer at a time, with continuous sutures.

An intramuscular injection (0.2 cc long acting penicillin) was given and the anaesthetic mask removed. A lateral view X-ray film was then taken of the left hind limb.

Once the rabbits were alert (approximately one hour), they were transported to Ellerslie. These animals were observed to resume normal levels of activity well within 24 hours post-surgery. All rabbits moved freely in their pens, weight bearing on the left hind limb when at rest and partial weight bearing with the ankle in plantar flexion when hopping around inside the pen.

The immobilisation period for all three groups was 57 days. Dates for immobilisation surgery were staggered over a six week period to allow sufficient time for testing. No more than three or four rabbits were tested on the arthrograph on any one day.

Pilot Studies

Over two years, fourteen different pilot studies were performed analyzing effects of a number of variables such as:

a. Sex of Animal Subjects

In previous research studies on the effects of joint-immobilisation, researchers tended to utilize rabbits of the male sex, although the experimental rationale for this was not mentioned.[5, 6, 9, 10, 11, 13, 14, 17, 253, 254] The present investigator found female rabbits easier to both handle and treat. In pilot studies, the males tended to 'thump' their hind limb when disturbed or frightened, an action that led to many femoral and tibial fractures in the immobilised limb.

It has been noted in the literature that, with rabbits, correct handling technique was necessary to prevent forced hyperextension of the lumbar spine, which could lead to serious injury.[194] Correct handling technique was made quite difficult with male rabbits. For instance, when the were removed from their cages, they tended to struggle vigorously to get free. By contrast, female rabbits seemed quieter and less aggressive during handling.

b. Form of Immobilisation

i) Steinmann pin with hook

This method was initially described by Woo et al.[254] A 2.4 mm Steinmann pin was inserted through the tibia and hooked over the anterior aspect of the femur (under the quadriceps muscle). An anterior tibial nut maintained the knee in a fully flexed position.

There were several drawbacks to this procedure. For instance, after insertion of the pin through the tibia, a hook was created by bending the pin with pliers. This bending levered a substantial force through the tibia and resulted in numerous tibial fractures. The majority of hooks also tended to slip off the femur by the second or third week. Moreover, the actual surgical procedure severely traumatized the quadriceps muscle while attempts were made to hook the pin over the femur.

ii) Steinmann pin with nut on only one end

This method was initially described by Akeson.[2] A Steinmann pin was inserted through both the tibia and femur. An anterior tibial nut maintained the knee in a fully flexed position.

Rabbits tended to be very active within their pens. Subjected to this frequent movement, the femoral aspect of the pin migrated from the femur, allowing more and more flexion-extension of the knee — to such an extent that the pin was often dislodged entirely from the femur.

iii) Nylon cord

A small diameter, multi-filament, nylon cord was used to hold the tibia and femur in full flexion. It was by means of a lateral incision that the cord was inserted, anterior to the femur and tibia, underneath the quadriceps and tibialis anterior muscles. The cord itself, however, tended to rupture after one or two weeks.

iv) External fixation of metal plates and threaded pins

This device proved too cumbersome for the rabbit to move; moreover, it easily caught in the mesh of the cage.

v) Plastazote splint

With their leg placed in a plastazote splint, the rabbits tended to gnaw at the exposed foot. Also, sores developed under the edge of the splint. Successful use of PVC-plastic splints has been reported in the literature, however subject rabbits' knees were placed in full knee extension.[130]

vi) Steinmann pin with two nuts, one at either end

This surgical immobilisation technique which was ultimately chosen produced minimal bleeding and trauma to the tissues of the rabbit's hind limb. Use of both anterior tibial and femoral nuts resulted in a particularly stable immobilisation. Incidence of fractures was minimal.

c. Diameter of Fixation Pin

During the first pilot studies, many femoral and tibial fractures were sustained in the early post-immobilisation stage. It was thought that the size of the Steinmann pin could be a factor in this overly-high incidence of fracture. Thus, pins ranging in size from 2.0 mm to 3.2 mm were tried, with a 2.4 mm diameter yielding the lowest incidence of post-surgical fractures. Pins larger than that size resulted in the highest incidence of fractures during surgery, and pins smaller than 2.4 mm broke (not the bone) within two weeks post-surgery.

d. Cage Dimensions and Flooring

Over the course of many pilots studies, different cages were used, varying in dimension from a width of 1.5 meters and length of 1.8 meters to the final dimensions of 50 cm x 50 cm x 50 cm. Cage size proved to be one of the most critical factors in reducing the incidence of fracture to an acceptable level. It appeared that the larger the cage dimensions were, the greater was the incidence of femoral and tibial fractures of the immobilised limb. Inside the larger cages, the rabbits were able to pick up speed whilst hopping and tended generally to engage in more gross movements. The possible effect of such gross movements in conjunction with the speed may have been an increase in stress applied to the bone, already weakened by the insertion of a pin. This weakening may well be significant in that Müller et al[154] have reported a 50 per cent reduction in bone torsional strength lasting from one to two months following insertion, or removal, of a single surgical screw within the diaphyseal region of the bone.

Any detrimental effect of cage size had been downplayed until a partic[illegible] time when the incidence of fractures seemed excessively high. A consulting veterinarian analyzed the problem as being the size of cage, not the surgical procedure (Wright M: personal communication, 1984).

In addition, problems with fractures (lower limb and lumbar) have been reported in past research involving rabbits where the cage flooring was wire mesh, such as in standard rabbit cages, and the rabbits' legs immobilised (Secord DC: personal communication, 1984). In the present study, for instance, if the immobilised limb became caught in the wire mesh, the rabbit frequently attempted to free the limb with sudden jerking movements. These movements often resulted in fractures. Once the cage flooring was changed from wire mesh to wood sawdust and shavings, the incidence of fractures and hind limb paralysis greatly decreased.

e. Environmental Factors

Rabbits are easily frightened by noise and the presence of other larger animals.[194] Even young rabbits, if frightened unduly and often, can die of heart failure (Secord DC: personal communication, 1984). Accordingly, the cages were placed in a quiet location away from the noises and presence of other animals.

f. Arthrograph

To provide more reliable results, the original arthrograph design was modified. The original fixation arms rigidly fixated the rabbit's hind limb within two metal beds and forced the knee to move in a simple arc with a fixed axis of rotation. This fixed axis was unlike the continually changing axis of the knee joint that normally shifts throughout its range of motion. The original rigid fixation forced an abnormal application of loads onto the knee joint and yielded an increase in measured knee friction.

The new limb support system allowed the tibia and femur to move independently of one another in response to any force placed upon it by the knee flexion-extension motion. Therefore, the ability of the arthrograph to isolate frictional force from other forces in the knee was further enhanced.

APPENDIX F

CALCULATIONS FOR FIRST CYCLE STIFFNESS SLOPES AND ENERGY

Calculations of 1st Cycle Stiffness Slopes.

The #33 immobilised knee will be used as an example for the determination of stiffness slopes (Figure III-10).

Machine calibration with the 500 gm weight was repeated six times prior to testing of the control knee and also six times prior to testing the treated knee (Figure III-8).

Average during #33 machine test = 58.7 mm deflection under 500 gm load.

Moment applied during calibration (same for all tests) = 462.4 Nmm (Figure III-9).

Calibration of vertical displacement from millimeters to Newtons:

$$= \frac{462.4 \text{ Nmm}}{58.7 \text{ mm}}$$

$$= 7.88 \text{ Nmm/mm}$$

$$= 7.88 \text{ N}$$

Therefore, the vertical displacement of 58.7 mm created by the 500 gm load was equivalent to 7.88 N.

Angular displacement (same for all tests) = 0.198°/mm. The slope of the 1st cycle to the point of linear load was calculated by dividing rise by run, or vertical displacement by angular displacement. As previously calculated in section 3.7c, the angular displacement for rabbit #33 was 81 mm and vertical displacement was 111 mm at the point of linear load.

For #33 immobilised Knee:

run = angular displacement to point of linear load on X axis

= 81 mm x 0.198°/mm

= 16.04°

rise = vertical displacement to point of linear load on the Y axis

$= 111\ \text{mm} \times 7.88\text{N}$

$= 874.68\ \text{Nmm}$

$= \dfrac{\text{rise}}{\text{run}} = \dfrac{874.68}{16.04°}\ \text{Nmm}$

$= 54.53\ \text{Nmm}/°$

Calculation of Energy

Energy = area (mm^2) x vertical displacement (N) x angular displacement (degrees/mm)

Using #33 immobilised 1st cycle as an example:

$$\text{Energy} = 21,299\ \text{mm}^2 \times 7.88\ \text{N} \times 0.198°/\text{mm}$$

$$= 21,299\ \text{mm} \times 7.88\ \text{N} \times 0.198°$$

$$= 33,231.55\ \text{Nmm}°$$

$$= 33,231.55\ \text{Nmm}° \times \frac{2\pi}{360}\ \frac{\text{rad}}{°}$$

$$= 579.1\ \text{Nmm (rad)}$$

APPENDIX G

RAW DATA

1

Areas of Hysteresis (mm^2) of Control Knees
for three groups over three cycles

Subject#	Group#	1st Cycle	2nd Cycle	5th Cycle
01	1	2,074	1,388	1,475
05	1	3,075	1,750	2,074
10	1	839	750	1,111
21	1	2:317	1,209	1,052
27	1	3,965	2,799	2,477
33	1	2,628	1,574	1,387
40	1	2,095	1,667	1,364
12	2	3,037	2,079	1,994
13	2	2,239	1,609	1,458
22	2	2,823	2,473	2,281
28	2	2,476	2,155	2,007
31	2	4,642	2,828	2,164
36	2	2,962	1,690	1,628
11	3	3,459	2,136	1,740
17	3	2,690	1,844	2,124
23	3	1,994	1,757	1,411
30	3	1,695	1,192	1,110
32	3	1,831	1,332	1,290
39	3	3,468	3,060	2,545

2

Areas of Hysteresis (mm^2) of Treated Knees
for three groups over three cycles

Subject#	Group#	1st Cycle	2nd Cycle	5th Cycle
1	1	16,518	2,466	1,732
5	1	33,054	2,590	1,366
10	1	19,447	2,497	1,272
21	1	10,320	1,199	956
27	1	16,148	1,427	849
33	1	21,299	1,268	1,034
40	1	11,313	2,479	703
12	2	18,211	5,044	3,148
13	2	30,440	5,355	4,166
22	2	10,264	3,148	2,252
28	2	26,262	4,686	3,571
31	2	14,718	1,476	1,042
36	2	22,515	2,064	1,059
11	3	13,525	2,503	1,919
17	3	5,277	2,732	2,000
23	3	13,515	1,757	1,129
30	3	9,421	764	537
32	3	14,462	2,445	1,645
39	3	12,517	1,193	653

(3) **Hysteresis Area Ratio 1st to 2nd Cycle for Treated and Control Knees**
of three groups

Subject#	Group#	Treated Area Ratio	Control Area Ratio
1	1	6.70	1.49
5	1	12.76	1.76
10	1	7.79	1.12
21	1	8.61	1.92
27	1	11.32	1.42
33	1	16.80	1.67
40	1	4.56	1.26
12	2	3.61	1.46
13	2	5.68	1.39
22	2	3.26	1.14
28	2	5.60	1.15
31	2	9.97	1.64
36	2	10.91	1.75
11	3	5.40	1.62
17	3	1.93	1.46
23	3	7.69	1.13
30	3	12.33	1.42
32	3	5.91	1.37
39	3	10.49	1.13

4

End-range Stiffness Cycles (mm) of Treated Knees
for three groups over three cycles

Subject#	Group#	2nd Cycle	5th Cycle	7th Cycle
1	1	25	21	19
5	1	62	60	56
10	1	40	38	37
21	1	17	17	17
27	1	15	15	15
33	1	19	18	17
40	1	23	23	22
12	2	22	22	20
22	2	27	25	25
28	2	20	21	21
31	2	21	20	20
36	2	36	36	36
11	3	31	28	27
17	3	32	32	31
23	3	22	21	20
30	3	13	12	12
32	3	29	29	28
39	3	33	29	28

⑤ **Mid-range Stiffness Cycles (mm) of Treated Knees**
for three groups over three cycles

Subject#	Group#	2nd Cycle	5th Cycle	7th Cycle
1	1	13	7	6
5	1	38	28	23
10	1	22	12	11
21	1	11	7	6
27	1	9	7	6
33	1	7	4	3
40	1	34	24	20
12	2	20	14	12
22	2	24	17	15
28	2	22	16	16
31	2	16	10	10
36	2	31	25	23
11	3	15	9	7
17	3	20	18	16
23	3	12	8	6
30	3	7	5	4
32	3	24	21	19
39	3	19	13	12

6 **First Cycle Stiffness Slopes (Nmm) of Treated Knees**
for three groups

Subject#	Group#	First Cycle Slopes
1	1	60.37
5	1	93.60
10	1	94.27
21	1	35.11
27	1	60.19
33	1	65.56
40	1	20.18
12	2	27.53
13	2	45.38
22	2	23.36
28	2	45.35
31	2	11.45
36	2	39.14
11	3	31.34
17	3	23.35
23	3	50.18
30	3	27.99
32	3	30.94
39	3	18.73

7 **First Cycle Vertical Displacement (mm) at Point of Linear Load**
for three groups of treated knees

Subject#	Group#	Vertical Displacement
1	1	55
5	1	121
10	1	138
21	1	50
27	1	69
33	1	111
40	1	116
12	2	100
13	2	177
22	2	84
28	2	136
31	2	86
36	2	127
11	3	94
17	3	91
23	3	81
30	3	35
32	3	101
39	3	102

(8) **First Cycle Angular Displacement (cm) at Point of Linear Load**
for three groups of treated knees

Subject#	Group#	Angular Displacement
1	1	6.7
5	1	8.2
10	1	15.2
21	1	9.0
27	1	6.8
33	1	8.1
40	1	18.8
12	2	18.7
13	2	20.4
22	2	24.2
28	2	13.7
31	2	20.7
36	2	18.8
11	3	19.8
17	3	25.2
23	3	15.8
30	3	8.8
32	3	24.2
39	3	23.3

⑨ **Ratio Vertical to Angular Displacement at Point of Linear Load**
for three groups of treated knees

Subject#	Group#	Ratio Vertical to Angular
1	1	0.821
5	1	1.476
10	1	1.021
21	1	0.556
27	1	1.015
33	1	1.370
40	1	0.617
12	2	0.535
13	2	0.868
22	2	0.347
28	2	0.993
31	2	0.415
36	2	0.676
11	3	0.475
17	3	0.361
23	3	0.513
30	3	0.398
32	3	0.417
39	3	0.438

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Present Status

1. Therapist-Owner, Alta-Orthopaedic Physiotherapy Inc., since January, 1986
2. Part-time Clinical Teaching Associate, University of Alberta, Faculty of Rehabilitation Medicine, Physical Therapy, since September 1980
   Instructor for PTHER 484: Assessment and Treatment of Joints, a fourth year physiotherapy course
3. Instructor of Post-Graduate National Orthopaedic Manual Therapy Courses, E1, E2, E3, V1, V2, V3 since 1977
4. Instructor, Advanced Vertebral-Extremity Anatomy Update, University of Calgary, since 1984
5. Instructor, Muscle Stretching Techniques, since 1984
6. National Examiner for the Orthopaedic Division of the Canadian Physiotherapy Association, since 1984

Degrees and Diplomas

1. Part B Canadian Manual Therapy Manipulation Examination (C.O.M.P.) Vancouver, July 1980
2. Part A Joint-mobilisation Diploma, International Seminar of Orthopaedic Manual Therapy, Gran Canaria, Spain, 1975
3. Joint-mobilisation Specialist, Oslo, Norway, 1975
4. B.Sc. (P.T.), University of Western Ontario, 1971

Professional Affiliations

1. Member of the Canadian Physiotherapy Association
2. Member of the following divisions of the Canadian Physiotherapy Association: Orthopaedic, Sports, Obstetrics, and Research
3. Member of the College of Physical Therapists of Alberta
4. Member of the Rehabilitation Engineering Society of North America

Grants and Awards

1. M.S.I. Foundation Grant, 1986
2. Operating Research Grant, University of Alberta, 1985
3. Alberta Recognition Award, Canadian Physiotherapy Association, Edmonton District, 1984
4. Congress Profits Fund Grant, Canadian Physiotherapy Association, Edmonton District

Related Work Experience

1. September 1983 - May 1985

    Part-time Clinical Teaching Associate, University of Alberta, Faculty of Rehabilitation Medicine, Physical Therapy

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3. December 1973 - June 1974

    Editor and translator into English from German, Manual Therapy for the Extremity Joints, for Freddy Kaltenborn in Gran Canaria, Spain and Oslo, Norway
4. February 1974 - December 1975

    Staff physiotherapist at Bydöy Allé Fysikalsk Institutt, Oslo, Norway

    Student at joint-mobilisation course, Oslo, Norway, instruction by Freddy Kaltenborn and Olaf Evjenth