

FEMUR FLAPS FOR TIBIAL RECONSTRUCTION: BIOMECHANICAL CONSIDERATIONS

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1. Femur Flap
2. Biomechanics
3. Fibula Flap

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Table of Contents

Table of Contents.....	2
Chapter 1: Introduction.....	9
Chapter 2: Background.....	10
Problem Description.....	10
Free-Vascularized Flap.....	11
Bone.....	12
Bone Material Properties.....	12
Structural Properties of Bone.....	15
Bone Response to Loading.....	16
Preservation of Bone.....	18
Bone Storage.....	18
Embalmmment.....	19
Materials Testing.....	20
Loading.....	20
Bone Simulation.....	22
Chapter 3: Project Approach.....	24
Chapter 4: Design.....	26
Problem Statement.....	26
Femur Flap Design.....	26
Experimental Design.....	27
Fibular and Femoral Flap Experimentation.....	27
Femoral Structure Experimentations.....	27
Chapter 5: Methodology.....	29
Specific Aim I:.....	29
Analysis of Test Results.....	33
Properties of Area of Flaps.....	37
Specific Aim II:.....	39
5.2 Compression Testing.....	39
Analysis of Test Results.....	43
5.3 Using Materials Testing to Construct Finite Element Models (FEM).....	44
Bone Quantities Used.....	45
Chapter 6: Data.....	46
Three Point Testing.....	46
Fibula Three Point Bending Testing.....	46
Initial Regional Studies of the Femur Harvest Region.....	47
Studies of the 35% and 40% Circumferential Femur Flaps.....	49
Compression Testing.....	52
Whole Femurs.....	52
Femurs After Removal of Flap.....	53
Finite Element Analysis.....	54
Chapter 7: Analysis and Discussion.....	57
Chapter 8: Conclusion.....	64
Chapter 9: Recommendations.....	65
Appendices.....	66
Appendix A.....	67

Appendix G.....	73
Appendix H.....	74
Appendix I.....	122
Appendix L.....	132
Appendix M.....	139
Appendix N.....	151
Appendix O.....	162
References.....	164

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Abstract

Femoral segments may reconstruct tibial fractures, substituting fibular segments. The femoral segment size superior to the fibular segment and the necessity for femur fixation were determined. In 3-Point Bending, a 40% diaphyseal circumference femoral segment failed 225N higher than the fibula ($p=0.047$). Using uniaxial compression and FEA, the 40% segment osteotomy initiated yielding in the femur at 2443N (safety factor = 2447N). As a result, the 40% femoral segment is superior to the fibula but requires fixation of the femur.

Table of Figures

Figure	Page	
2.1	Area of Vastus Intermedius Insertion from the Anterior View	10
2.2	Stress-Strain Curves for Human Femur, Tibia, and Fibula	16
2.3	Bending Loads	20
2.4	Bending Loads in a Femur	21
5.1	An Illustration of the 3 Point Bending Construct	29
5.2	Theta	32
5.3	Flap Geometries	33
5.4	Free Body Diagram of 3-Point Bending	34
5.5	Internal Moment	34
5.6	Radius of Curvature	36
5.7	Photo Analysis	37
5.8	Parallel Axis Theorem	38
5.9	Femur Orientation	40
5.10	An Illustration of the Compressive Loading on the Femoral Specimen	41
5.11	Positioning of Strain Gauges around the Donor Site	42
6.1	Force Versus Displacement Curve of a Fibula Flap in 3-Point Bending	46
6.2	Force Versus Displacement Curve of Regional Studies	48
6.3	Force Versus Diaplacement of 35% Femur Flap	49
6.4	Force Versus Diaplacement of 40% Femur Flap	50
6.5	Force Versus Diaplacement (35% Vs. 40%)	50
6.6	Femurs Versus Fibula Flaps in Three Point Bending	51
6.7	Full Femur Compression	52
6.8	Force-Displacement Curve of a Femur with a 35% Flap Removed	53
6.9	Compression Study of the Femurs	54
6.10	Force-Displacement Curve from FEA	54
6.11	Femur Model Prier to Failure	55
6.12	Femur Model at Failure	56
7.1	Average Stress-Strain Curve of Fibula Flaps	58
7.2	Average Stress-Strain Curve of Femur Flaps	59
7.3	Average Stress-Strain Curve for the 35% and 40% Femur Flaps in Comparison to the Fibular structure	60
7.4	Lever Arm	62

Table of Tables

Tables	Page
2.1 Bone and Cement Properties for Finite Element Modeling	14
6.1 Resultant Maximum Loads and Yielding Loads of All Fibula Flaps	47
6.2 Resultant Maximum Loads When Determining Circumferential Percentage	48
6.3 Resultant Maximum Loads of 35 and 40 Percent Femoral Flaps	51
6.4 Resultant Maximum Loads in Whole Femurs	52
6.5 Resultant Maximum Loads in Femur with 35% or 40% Flap Removed	53

Chapter 1: Introduction

University of Massachusetts Medical Center has proposed a new procedure that would be used to reconstruct a non-continuous tibial fracture of six centimeters and longer. The novel procedure employs the use of a femur free vascularized flap to replace the function of the fibula free vascular flap in the reconstruction of a tibia. To further progress in the development of the surgical procedure, UMass plastic surgeons need to determine the dimensions of the femoral flap that will not compromise the structural stability of the femur excessively. The Worcester Polytechnic Institution MQP team will evaluate the structural stability of the donor site in the femur as well as the structural stability of the femoral flap itself. The femur flap structure will be evaluated and compared to the structural stability of the current fibular flap. From this analysis the MQP team will decide which flap provides more structural support in the tibia. Once the MQP team makes their evaluation, then the UMass Medical School can further progress on the development of their surgical technique.

This report illustrates the process followed to accomplish the Major Qualifying Project's goals. The background research, the methodology of the teams work, the test results and the analysis of the data will be presented to show the path the team took to arrive at their conclusions.

Chapter 2: Background

Problem Description

Plastic surgeons at the University of Massachusetts Medical Center have proposed a new procedure to reconstruct a tibia defect of six centimeters and longer. The procedure involves the use of the insertion area of the vastus intermedius (Figure 2.1) of contralateral femur as a donor site rather than the frequently used fibula.

Figure 2.1:

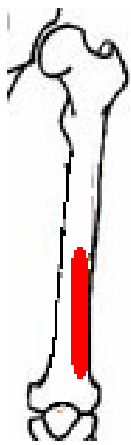


Figure 2.1: Area of Vastus Intermedius insertion from the anterior view (adopted from www.meddean.luc.edu).

After the site has been cleaned and the defected bone is properly shaped to accept the bone graft, the free vascularized flap of the femur will be implanted. Micro-surgeons then reconnect the vessels of the flap to the local vascular system surrounding the location of the defect. The patient will be observed to determine when osteointegration and bone growth develop. Progressively, the doctors will prescribe the appropriate loading when sufficient bone fusion and growth takes place; however, initially the patient will be permitted to apply no loading to the reconstructed limb.

The plastic surgeons are most concerned with the circumferential percentage of the femur that may be removed without compromising structural stability of the donor site in withstanding the patient's body weight. The patient will have the reconstructed leg immobilized for the initial healing period, but will have making the patient depend on the contralateral femur for some mobility. However, once the defect site has adequately healed, the patient will be advised to apply gradually increasing loads to the reconstructed tibia in order to encourage further bone

growth. Furthermore, plastic surgeons are concerned with the structure stability of a cortical cylinder configuration of the fibular flap and that of the wedge configuration of the femur flap.

The flap will be removed from the vastus intermedius origin on the anterior distal lateral region of the femur. The study requires the measurement of what radial percentage of the femur may be removed without significantly affecting its structural stability. This flap is to be implanted into the tibial defect.

The femur flap will be structurally analyzed in comparison to the commonly used fibula flap. This analysis will be used to determine if the femoral flap will provide the same support as a fibular flap. Furthermore, if it is necessary, a larger segment of the femur could be used to mimic the structural support of the fibula. The weakened femur structure would be reinforced using an Intramedullary Nailing System. The analysis of this graft structure will aid in determining a surgical technique that will maintain the stability of the complex bone structure of the femur.

Free-Vascularized Flap

In the reconstruction of the tibial defect, a free-vascularized flap will be used. A free-vascularized flap is a segment of tissue that has been fully removed from its donor location with a system of intact blood vessels. The term “free-vascularized flap” may pertain to the use of skin, muscle, and bone. When a free vascularized bone flap is removed from the donor site, skin and muscle are often kept attached to help maintain the vascular system that feeds the bone segment (Soutar, 1994). Two types of blood vessels may supply the bone segment. One type of supplemental vasculature is the nutrient vessels (septal branches that vascularize both the muscles and the skin) that feed remote locations of the bone. Microvascular surgeons would need to use the larger vessels found in the muscle and or the skin, which are a part of the flap, to maintain blood supply to these nutrient vessels. Another type of supplemental vasculature would be a bone perforating vessel that is much larger than the nutrient vessels. This blood vessel supplies a much larger area of the surrounding tissue. Microvascular surgeons can use these perforating vessels to give the bone flap a blood supply at the graft site (tibial defect) (Soutar, 1994). Grafted bones with a surgically implanted blood supply stand a much higher chance of survival than bone grafts that do not have a direct blood supply (Soutar, 1994).

Other surgical techniques have been used to reconstruct tibial defects that are non-continuous and range from six centimeters and longer. The team needed to be aware of other

procedures that the new procedure being tested could be compared against. In this section the team describes the current technique of using vascularized fibular grafts to reconstruct the defect. To reconstruct a defect of the tibia six centimeters and larger, a common technique of vascularized fibula graft is used. This process involves using a large segment of the fibula bone, which will be formed to fit the defect in the tibia. The segment of bone has an intact vascular system that will maintain a supply of nutrients to the bone graft. The ends of the tibial defect are shaped to better accept the bone graft. The fibula graft remains as a cylindrical bone with the ends of the bone being surgically removed. The use of internal and external fixations techniques depends on the situation of the fracture.

This surgical technique has been effective in saving the limbs of patients that have large non-continuous defects in their tibia. The technique, however, does have morbidity problems for the donor-site, and complications of stress fractures of the grafted bone (Enneking, 1980). Donor-site morbidity involves hypersensitivity at the donor-site and sensory loss in the foot, as stated by the summary of the project presented by UMass medical. These stress fractures require the attention of the doctor who may require the patient to undergo another surgery for external immobilization, or for another bone graft for the un-united fracture (Enneking, 1980).

Bone

Bone Material Properties

Various factors affect the integrity of bone, especially during preparation of the test specimen. The variability is due to the type of bone, trabecular or cortical, the age, disease state, and bone source, and the experimental environment, including temperature, specimen hydration, and preservation. Immediate proper care of the removed bones greatly affects the resulting strength of the bones. When bone is dried it becomes more brittle and its strength and Young's modulus increase due to the increase in stress needed to cause fracture. Literature shows that drying the bone results in a 31% increase in ultimate tensile strength and a 55% decrease in toughness (Burr, 1993).

One way to decrease variables that may affect experimental results is to store bone in an environment that closely resembles its native environment. This is accomplished by storing the test species in physiological saline and then wrapping them in saline-soaked gauze during testing. Strain rate, also, needs to be considered. Dried bone acts like a spring whereas the wet bone acts like a spring with "shock absorbers" (Burr, 1993). When imitating the physiological

conditions, strain rate should be set at the range that occurs in vivo, between 0.01/s and 0.08/s (Burr, 1993). Refer to Table 2.1 for the table of material properties of wet bone in the femur, the tibia, and the fibula.

Table 2.1. Bone and Cement Properties For Finite Element Modeling

	CORTICAL BONE		POTTING CEMENT	TRABECULAR BONE
	Longitudinal	Transverse	PMMA	
Comp. Yield Strength (MPa)	182	121	68.95-131*****	20.6+6.5**
Comp. Ult. Stress (MPa)	195	133		1.5-50*****
Comp. Ult. Strain (%)	2.2-4.6			1.9
Compressive Yield Strain (%)	1.9			0.88 ± 0.06**
Compressive Modulus (GPa)			2.5-3.1*	0.1-3*****
Tensile Yield Stress (MPa)	115			13.8+4.8**
Tensile Ult. Stress (MPa)	133	51		3-20*****
Tensile Ult. Strain (%)	2.9-3.2			
Tensile Yield Strain (%)	1.18			0.41 ± 0.04**
Elastic Modulus (GPa)	9.6	17.4	3.5*	18.6+2.2**
Shear Ultimate Stress (MPa)	65-71****			6.6*****
Shear Modulus (GPa)	3.6*****	3.3*****	802.5-1365.2*****	
Poisson's Ratio	0.58***	0.31***	0.384-0.403*****	
Ult. Shear Axial (MPa)				9.8(6.1)*****
Ult. Shear Radial (MPa)				9.5(5.9)*****

SOURCE:

Martin et al 1998

* Modern Plastics Encyclopedia 1999, from Roger D. Corneliussen 2002

**Bayraktar et al 2001

***Reilly, D.T. and Burstein, A. H., The elastic and ultimate properties of compact bone tissue, J. Biomechanics , 8, 393-405, 1975.

****Van Buskirk, W. C. and Ashman, R. B., The elastic moduli of bone, in Mechanical Properties of Bone , Joint ASME-ASCE Applied Mechanics, Fluids Engineering and Bioengineering Conference, Boulder, CO, 1981.

*****Garnier et al. (Mechanical characterization in shear of human femoral cancellous bone: (torsion and shear. 1999

*****Yoon, H.S. and Katz, J. L., Ultrasonic wave propagation in human cortical bone. II Measurements of elastic properties and microhardness, J. Biomechanics , 9, 459-464, 1976.

*****<http://www.orthoteers.co.uk/Nrujp-ij33lm/Orthbonemech.htm>

*****CES Selector, 2004 by Grant a Design Limited

Structural Properties of Bone

Bones are loaded under various conditions including tension, compression, torsion, and bending. These conditions determine the formation of the bone; the bone is constructed for certain loading. Failure depends on the type and the direction of loading. For example, when a compressive force is applied to bone, the bone buckles and fails in shear because of the 45° angle that the shear forms with the compressive force. Compressive stresses develop when a force is applied such that it compresses, or shortens, the bone. Tensile stresses exist when the bone is stretched. And shear stresses exist when one region slides relative to a neighboring region within the bone. Deformation in a bone's length alters the width of the bone and Poisson's ratio is the ratio of width strain to length strain (Burr, 1993).

Failure is described as the region outside of the elastic region on the load-deformation curve known as yield point. This is the failure region because there exists a small post-yield strain, or material ductility, in bone prior to fracture (Burr, 1993). The femoral head is able to withstand loads up to 35% of body weight when standing. The shaft of the femur can withstand axial compression forces and bending moments up to 40% of body weight. When the femur bends under axial compression, the lateral surface of the femur is in tension and the medial surface is in compression. The femur is attached laterally to the hip at an angle of 45°. The area under the stress-strain curve represents a material's toughness, which relates the resistance to fracture. The maximum stress that a bone can sustain is referred to as the ultimate strength, while the breaking strength refers to the stress at which the bone breaks. Refer to table 2.1 for the material properties of the bones relevant for this experiment. In testing, the structure of bone is assumed linearly elastic in order to relieve some of the complicated stress analysis that would exist otherwise (Burr, 1993, Yamada, 1970).

Figure 2.2: Stress-Strain Curve for Human Femur, Tibia, and Fibula

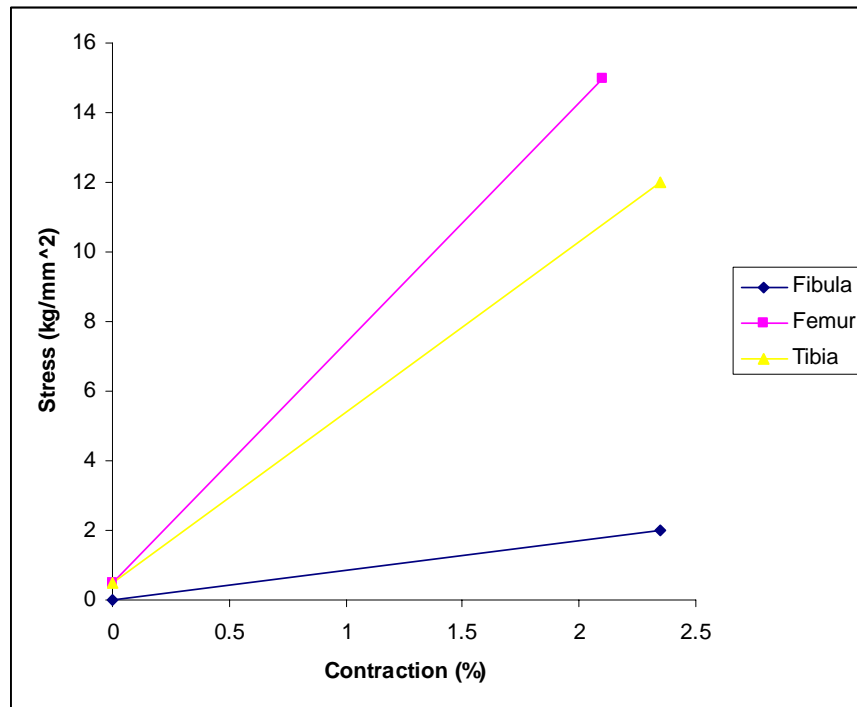


Fig. 2.2: The femur, tibia, and fibula are very strong yet brittle and thus, there exists very little deformation outside of the linearly elastic region for each bone before the respective bone breaks (based on Yamada, 1970).

The slope along the elastic region in the load-deformation curve represents the rigidity of the structure. Larger bones tend to be more rigid. The elastic region along the slope in the stress-strain curve represents the Young's Modulus. In cancellous bone, the material's stiffness is that of a single trabecula and the structure's stiffness is that of the trabecular structure as a whole. Structural properties are proportional to cancellous bone density and the arrangement of trabeculae (Yamada, 1970). The hardness value of the tibia is 0.375GPa, of the femur is 0.35 GPa, and of the fibula is 0.22GPa (Yamada, 1970). Thus, it can be seen that the tibia is the stiffest and the fibula is the softest. Cancellous bone can withstand strains up to 75% before failure, while cortical bone fractures at a 2% strain (Bindal, 2002).

Bone Response to Loading

Compressive tests push the ends of test samples together by applying a uniaxial compressive load. The compressive stress is calculated using the formula: $\text{stress} = -F/A$, where F is the axial force and A is the cross-sectional area. The specimen is loaded in the test machine

using abutments placed at the ends of the bone which are each attached to the force transducer. However, if the bone is not properly lined up then large stress concentrations can exist at the ends of the bone and failure would occur sooner than it would if it were in anatomical loading. When aligned correctly, compression tests simulate in vivo loading conditions of the bone, where one-third of failure is as a result of tension (Burr, 1993; Fung, 1981).

Compression tests allow for testing in a system that simulates the weight bearing on bones under static conditions within the human body. It allows one to find maximum compressive loads that can be placed on each individual bone without interfering with bone's integrity.

Bending tests load the bone in bending until failure. Stresses in bending are calculated using the formula: $\text{stress} = Mc/I$, where M is the bending moment, c is the distance from the center of mass of the cross-section, and I is the moment of inertia of the cross-section. Bending creates tensile stresses along one side of the bone and compressive stresses along the other. Bone is weaker in tension than in compression and, thus, failure will occur on the outside, or tensile side, of the bone where the greatest stresses exist (Yamada, 1970; Bindal, 2002).

There exist many types of bending tests: such as three point and four point loading. The bone must be long enough, as compared to the width, to show correct results of bending; because if it is too short then the displacement will be due to the shear stresses and not to the bending. Three point bending systems create shear stresses near the bone's midsection while four-point bending creates pure bending. However, four-point bending is difficult when testing irregular shapes because the force at each loading point has to be equal in order to create pure bending. In both bending tests there exist deformations in the bone where the load is applied to the surface (Burr, 1993).

Bending tests simulate the weight bearing on bones under bending conditions within the human body, which allows one to find maximum bending loads that can be applied to bones without causing fractures.

Tensile tests stretch the bone and measure the resulting strain. Most of the failure is due to tension in tensile stresses. However, in order to test in tension and simulate the response under anatomical conditions, the specimen has to be large, which allows the structure to be treated as a continuum (Burr, 1993).

Tensile tests simulate the tension placed on bones under static conditions within the body. It allows one to find maximum tensile loads that can be placed on each individual bone without causing rupture.

Combined loading tests load the specimen in compression and bending within the same test. Buckling will occur due to shear stress before crushing occurs along the axis of the bone. The stress across the cross-section of bone is calculated using the formula:

$$\sigma = (-F/A) \pm (My/I),$$

where M is the bending moment, y is the distance from the neutral axis, and I is the area moment of inertia. Combined loading tests simulate the bending due to compression placed on bones under static conditions within the human body. It allows one to find maximum combined loads that can be placed on each individual bone without causing buckling (Bindal, 2002; Burr, 1993).

Preservation of Bone

To clearly understand experiential data, the effects of preservation techniques used to keep the bone material from degrading were researched. The bone specimens that are to be supplied by the University of Massachusetts Medical School will be obtained from cadavers. These cadavers have been chemically fixed with formaldehyde to prevent decomposition of the bone structure. The cadavers also had to be embalmed before the school received them. The effects of the embalming on bone mechanical properties were researched to understand the mechanical response of the chemically fixed bone and make estimations as to how living bone would respond to the same loads that are applied in the experimental part of this research. Furthermore, proper storage technique is required to maintain bone during the experimental portion of the research.

Bone Storage

To maintain the properties of bone once it has been removed from the cadaver, the bone sample will be kept moist in a saline solution. To keep the sample moist saline solution will be applied to the bone through the use of gauze that has been wrapped around the bone. Once the bone is wrapped in the moistened gauze, the bone and gauze will be placed into a plastic bag. The plastic bag with the bone will then be placed into an environment of -20 degrees C (Cowin, 1989).

The affects on the material properties for storing of the bone sample using this treatment were researched to determine what changes may be observed in testing. It has been shown that the lower temperature has no effect on the mechanical properties of long bone under torsional and four-point bending test (Goh, 1989). Screw pullout tests have shown that low temperatures do not affect cancellous bone (Matter, 2001). The cancellous bone was also shown not to have any change in stiffness through many thawing, testing and refreezing sequences; however the viscoelastic properties did show to have a small sensitivity to this handling (Linde, 1993). Cortical bone compression is shown not to have any significant difference when treated with liquid nitrogen, for treatment of bone tumors, compared to the natural bone (Yamamoto, 2003). It is also been found that to keep the material properties of the bones it is important to keep the bone frozen at temperatures around -20 degrees C (Cowin, 1989).

Embalmmment

A common method of preserving human bone specimens is through the process of embalmmment. This process requires the specimen to be immersed in a series of 95% alcohol, 10% formalin (formaldehyde), and pure glycerin, which is identical to the fixation process experienced by the supplied bones. Due to the fact that the bone samples will be taken from cadavers at the University of Massachusetts Medical School Morgue, the specimen will have went through an embalmmment process as discussed earlier to maintain preservation.

Such methods of preservation have an effect on the mechanical properties of the cadaver's tissue. More importantly, it affects the mechanical properties of the bone specimen. Calabrisi et al. determined that the embalmmment process decreases the compressive strength of the femur by 13%. However, these results did not have statistical significance with $p > 0.12$ (Calabrisi, 1951). Other studies have shown that the elastic modulus of a femoral segment has an Elastic modulus of 3.92 GPa when fresh and 4.09 GPa after embalmmment in 10% formalin. However, the data was again not statistically significant at $0.1 > p > 0.05$ (Sedlin, 1965). The final experimentation was conducted on human male tibial segments. It was observed that the Elastic modulus increased by 16.3% from 2.14×10^6 psi to 2.49×10^6 psi ($p < 0.01$). The Ultimate Tensile Strength (UTS) increased by 10% from 12,290 psi to 13,514 psi ($p < 0.01$), and the Elongation Percentage decreased by 20.6% from 1.959% to 1.555% ($p < 0.01$) (Evans, 1973).

In summary, the embalment process has an insignificant effect on the mechanical properties of human bone. The data previously described shows that the Elastic modulus increases, the UTS increases, Elongation % decreases and the compressive strength decreases as the bone is fixated using the embalment process.

Materials Testing

Loading

A bending moment is created by the asymmetrical geometry of the femur and the nonlinear loading asymmetrical compressive load applied by the acetabulum and the tibia. Such bending leads to the creation of combined loads, where multiple types of loads are applied to the same body. In the case of the femur, it experiences compressive and bending loads externally, but also experiences compressive and tensile loads internally. In testing the femur, the positioning of the femur is critical as it creates a bending moment and initiates different internal loading as shown in Figures 2.3 and 2.4.

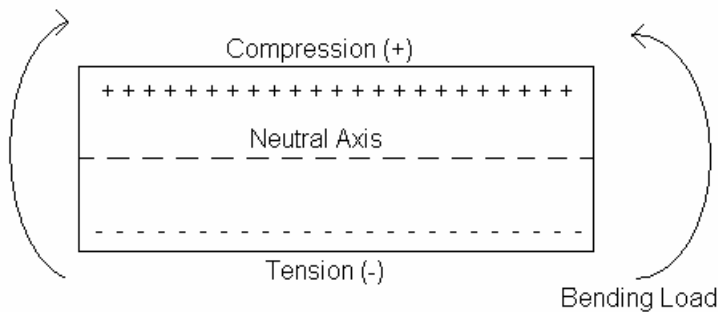


Figure 2.3: Bending Loads - A bending load creates tension on one side of the specimen and creates compression on the opposite, the neutral axis experiences no forces because the compressive and tensile force are equal and negate each other.

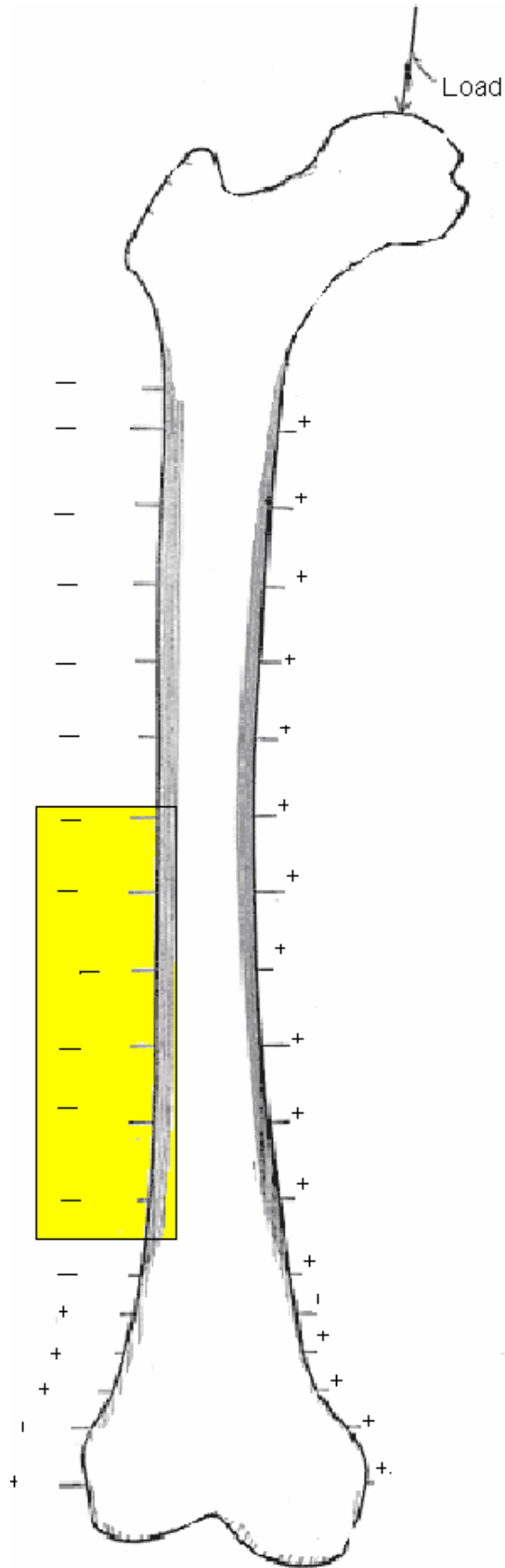


Figure 2.4: Bending Loads in a Femur- This drawing illustrates the stresses that are experienced in the femur while being loaded. The illustration shows that the medial diaphysis experiences compressive loads (+) and the lateral diaphysis experiences tensile loads (-). The anatomical positioning of the femur creates these internal stresses. The angled positioning and asymmetric geometry creates a bending moment. The region from which the proposed femoral flap will be removed experiences tensile stresses as seen in yellow (adopted from Bartleby.com).

To ensure life-like loading, a method commonly used for holding a whole bone in a materials testing device is referred to as “potting”. Heiner et al used this method where the bone is held in place by a cup at the femoral condyle and the femoral head. The cups are filled with Polymethylmethacrylate (PMMA) to transfer the load being applied to the fixation device to the bone in the appropriate location and direction. This will insure life like loading scenarios in the experimental testing of the femur (Heiner et al, 2001).

Bone Simulation

The supply of cadaver bone specimen for experimental procedures is limited and therefore steps must be taken to efficiently use the bones for experimentations. In order to take advantage of the specimen, simulation of the bone deformation under loads will aid in the search for appropriate load applications and flap dimensions. There are several methods of simulating the bone structure and its response to loads when geometric parameters of the structure are altered. The methods that are being considered are computer analyses and possible composite bone models.

Finite Element Analysis (FEA) is a technique for predicting the response of structures and materials to environmental factors such as forces. The process starts with the creation of a geometric model. The model is subdivided into a mesh composed of small elements of simple shapes connected at specific node points. In this manner, the stress-strain relationships are more easily approximated. Finally, the material behavior and the boundary conditions are applied to each element. FEA is most commonly used for structural and solid mechanical analysis where stresses and displacement in response to loading are calculated. These parameters are often critical to the performance of the structure and are used to predict failures (Structural Research & Analysis Corporation, © 2003). There are many programs that offer structural and dynamic analysis of loading effects. The three being considered for this experimental research are ABAQUS®, ANSYS®, and LS-DYNA®.

ABAQUS, Inc. produces a Finite Element Analysis program by the name of ABAQUS/Standard version 6.4, which is specifically designed to analyze linear and nonlinear modeling. This option allows for the study of structures such as human bone that displays a quasi-linear viscoelasticity response to loading (Lakes, 1979). Furthermore, the program is capable of performing nonlinear static and nonlinear dynamic analyses. A biomedical

application to which this product has been previously used for is the analysis of Nitinol material implemented in intravascular stents. The properties that were analyzed were the fatigue strength of the material under cyclic loading created by the pulsatile blood flow and the cyclic expansion and contraction of the blood vessel (ABAQUS, Inc. © 2003, 2004).

ANSYS, Inc. offers an FEA program by the name of ANSYS Mechanical®, which specializes in nonlinear simulation. Specifically, the program contains comprehensive Maxwell and Kelvin models that are used to model the viscoelasticity of tissue (Bhashyam, 2002). A biomedical application to which this program was applied was the simulation of total hip replacement surgery. The ANSYS FEA program was used to simulate the nonlinear tissue response to press fitted acetabular implants. Furthermore, the program was used to reproduce the complex structure of the joint and the implant materials (DiGioia III, 1996).

In order to use these program options, the geometric parameters of the bone model must be set. Models that have these geometric parameters have previously been created and used by researchers, and are deposited at the Biomechanics European Laboratory Repository where M. Papini and P. Zalzal have deposited a 3rd generation composite femur for ANSYS programs.

LS-DYNA is used to simulate events over an interval of time and the deformations that result. Dynamic loading is used to solve for non-linear structural properties. LS-DYNA allows for simulation of 2-D and 3-D elements within a structure, and single-surface, surface-to-surface, and node-to-surface contacts. It allows for simulation of anisotropic materials, such as bone, which would give more accurate results in testing the structural strengths of the tibia, femur, and fibula because it would not have to be assumed an isotropic material in order to simplify stress analysis (<http://ansys.com/>, 2002).

Chapter 3: Project Approach

This MQP project is a scientific project that attempts to validate whether or not the femur flap is a superior structure than the traditionally used fibula flap in tibial reconstruction. The doctors in the Plastic Surgery division at UMass Medical proposed the vastus intermedius origin on the femoral diaphysis (distal anterolateral) as a source of a large bone segment that may be used as a free-vascularized flap. The plastic surgeons, however, are not knowledgeable with respect to the size of the femoral segment that is needed to simulate or exceed the fibular flap. Furthermore, the effects on the femur after the removal of such a femoral segment are unknown. The doctors have empirically shown that 30% of the circumference can be removed from the radius (forearm bone) before the radius is weakened by 25%. Such assumptions cannot be associated with the femur as it is a weight bearing bone and an osteotomy may have catastrophic effects.

We hypothesized that an adequately sized free-vascularized femoral flap may be removed from the femur without exceeding the elastic limit of the femur under anatomical compressive loads. Furthermore, the femoral flap will tolerate a greater maximum load, in 3-point bending, before failure as compared to the traditional fibular free-vascularized flap.

To test this hypothesis the project was divided into two specific aims. The first specific aim was to determine the minimal circumferential percentage of the femoral diaphysis that will provide a stronger segment than the fibular flap. This was done through comparing the circumferential percentage that is necessary to exceed the fibular flap in maximum load by 100N. The second specific aim was to determine the structural integrity of the harvested femur. The harvested femur must not exceed its yield strain while being loaded under fiftieth percentile male body weight during stair descent after the minimal circumferential percentage is removed.

The designed experiment to test specific aim one, involves the use of human fibula flaps and multiple sized (changes in percent circumference) segments of human femur bone, the whole circumference is used where the femur flap is to be removed for initial studies. The fibula flaps and femur segments are subjected to 3-point bending, as this tests the bones in their weaker material properties by creating a bending moment and tensile forces. The femur segments are compared to the fibula flaps to determine what sized segment best matches and exceeds the fibula flap in maximum load, so the fibula flaps tests were first. Next, the determined segment was removed from the femur at the vastus intermedius origin (distal anterolateral region of the

diaphysis) as proposed by plastic surgeons based on vasculature availability. Segments of 15% to 35% diaphyseal circumferences (in 5% increments) were used to determine the size of the femoral segment that will match the maximum load of the fibula flap. Thirty-five and forty percent segments were then harvested from the femur to determine the how much higher of a maximum load a 40% segment can tolerate. Only two segment sizes were used as to collect enough data to perform statistical analyses.

The designed experiment to test specific aim two, utilized the whole human femurs as well as femurs with 35% and 40% segments harvested. These femurs were placed in anatomical position under uniaxial compression device to simulate natural occurring forces in the body. Whole femur maximum loads were compared to those of the harvested femurs. Harvested and whole femurs had strain gages attached in similar locations to analyze the weakening of the femoral structure as the harvested segment size increases. This strain data was used in finite element modeling to create the femur model that will simulate the femurs used in the experimental testing. Using this finite element model, the load at which the femur exceeded the elastic limit was determined. This load was compared a developed safety factor load (stair descending fiftieth percentile male). Furthermore, whether the removal of a 35% or 40% segment weakens the femoral structure so greatly that it exceeds its elastic limit prior to the establish safety factor load. Such a result would indicate that restructuring of the femur is required.

Chapter 4: Design

This project consisted of both scientific and engineering design. The predominant focus of this project was to design a femoral segment for the reconstruction of discontinues tibial fractures. In order to assess the femoral flap design, an experimental procedure was designed. In this project, plastic surgeons at UMass Medical Center and biomechanical specialists were regarded as the stakeholders of our designed femoral segment. The plastic surgeons were the primary stakeholders as they would be using this project's results to further develop their surgical technique for tibial reconstruction. Biomechanical specialists were the secondary stakeholders as they would refer to our results for future analyses. Thus, the experimentations must be valid in methods that are currently used in the biomechanical/orthopedic field.

Problem Statement

Traditional free-vascularized fibular flaps are associated with high risks of post-operative graft fracture and arterial thrombosis (Arai et al., 2002). To avoid these complications, plastic surgeons have proposed the use of a free-vascularized femoral flap from the origin of the vastus intermedius muscle for the reconstruction of discontinuous tibial fractures of six centimeters and longer (Dunn et al., 2003). The surgeons need to know what size femoral flap is required to be superior than the fibular flap in prevention of post-operative fractures, as well as, how much the donating femur is structurally compromised.

Femur Flap Design

The primary objective of this project was to design a femoral flap for the reconstruction of discontinuous fractures. In the design of the femur flap, a weighted objectives chart was completed. The chart accounted for the project advisor's (biomechanical expertise) and surgical advisor's (clinical expertise) opinions to determine what objectives the femur flap should satisfy. For analysis, the surgical advisor's opinion was weighted with 70% as they will be the clients that will be using this surgical procedure on a regular basis. The opinion of the project advisor was weighted as 30% (Appendix A). The following is a listing of some of the objectives in ranking of importance:

The Femoral Flap must...

- 1st ...be removed from the vastus intermedius origin on the femur
- 2nd ...not compromise the structural integrity of the femur upon removal
- 3rd ...withstand higher loads than fibula
- 5th ...fit to current osteotomy techniques used by surgical clients
- 6th ...be effective in reconstruction of tibial fractures > 6 centimeters

Due to constraints discussed in the methodology, the femoral flap was designed so that it may be removed from the vastus intermedius origin on the femur, fits current osteotomy techniques, and withstands higher loads than the fibula.

Experimental Design

Fibular and Femoral Flap Experimentation

To design an effective experimental method of comparing fibular and femoral flap material properties, the following experimental options were assessed:

- Shear Tests
- Compression Tests
- Tensile Tests
- 3-Point Bending Tests
- 4-Point Bending Tests
- Combined Shear-Compression Tests
- Combined Shear-Tensile Tests

These experimental techniques were evaluated in their capacity to satisfy requirements (i.e. use occurrence by previous experimentations in the biomechanical field and simulation of natural loading) using a Pairwise Comparison Chart (Appendix B). Once it was established that the 3-Point Bending Test would be the optimal experimental technique, a method of comparing testing hardware designs was also completed. The Pairwise Comparison Chart concluded that the 3-Point Skid Design was the best design choice for performing 3-Point Bending Tests on fibular and femoral flaps (Appendix C). (Refer to Appendix D for hardware images)

Femoral Structure Experimentations

To design an effective experimental method of assessing the effect a flap removal has on the femur structure, the following experimental options were assessed:

- Dynamic Loading
- Compression Test Along the Diaphyseal Axis
- Compression Test in Anatomical Orientation
- 4-Point Bending
- Tension Test Along the Diaphyseal Axis

These experimental techniques were evaluated in their capacity to satisfy requirements (i.e. use occurrence by previous experimentations in the biomechanical field and simulation of natural loading) using a Pairwise Comparison Chart (Appendix E) . Once it was established that the Compression Test in Anatomical Orientation would be the optimal experimental technique, a method of comparing testing hardware designs was also completed. The Pairwise Comparison Chart concluded that the *Fixed Free Tree Stand Design* was the best design choice for performing compression test in anatomical orientation on the femoral structures (Appendix F). (Refer to Appendix G for hardware images)

Chapter 5: Methodology

All bones were wrapped in saline-soaked gauze, placed in plastic bags, and kept in a freezer upon removal from the cadavers in order to preserve the bones. Twenty-four hours before the bones were used in the experimental methods, the corresponding specimens were placed in a fridge at 4° C to allow them to thaw.

Finite Element Analysis

Specific Aim I:

The first specific aim was to determine the minimal circumferential percentage of the femoral diaphysis that will provide a stronger segment than the fibular flap. This was done through comparing the circumferential percentage that is necessary to exceed the fibular flap in maximum load by 100N.

5.1 Three Point Bending

The 3-point bending attachments, which were manufactured by Steve Derosier in the Washburn Machine Shop, were installed onto an MTS 858 Bionix® (Biomaterials Laboratories, Mechanical Engineering Department, Worcester Polytechnic Institute). Refer to Appendix D for the design of the attachments. The bone specimen was situated on the abutments so that the lower abutment supported the bone at 10% and 90% of the length of the bone as seen in figure 5.1. The upper abutment was lowered onto the specimen at a strain rate of 0.01 mm/sec so that the force/strain data could be acquired precisely (Norrdin, 1995).

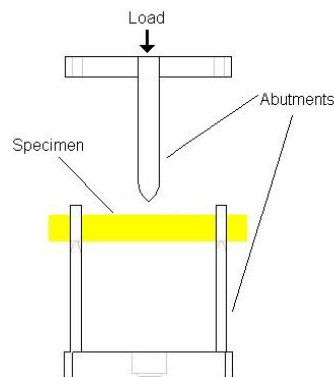


Figure. 5.1: An Illustration of the 3 point bending construct

- 5.1.1 A cylindrical piece, which had a length of 10cm, was removed from the fibula. The flaps were measured, 10cm in length, and removed from the middle of the shaft of the fibula using an oscillating orthopedic saw. The bone was continuously sprayed with saline while sawing to prevent the saw from burning the ends of the bone and to keep the bone wet. The removed flaps were wrapped in saline-soaked gauze to prevent them from drying out while waiting to be tested. Three point bending was applied to a fibula flap in order to determine its strength. This was repeated for thirteen fibula samples. Refer to Appendices J and K for the list and description of the bone specimens used. The grafts were loaded until failure, which was when the bone cracked, in order to determine the strengths of the different flaps.
- 5.1.2 Originally, our MQP group was going to use four whole femurs in compression testing, as mentioned in 5.2.4, to validate the FEM for Luca et al. This, in turn, would allow Luca to remove sections of the mesh, which would represent removal of the flap from the femoral structure. Three-point bending of cylindrical fibula flaps, with a set length of 10cm, was going to be simulated in FEM, by Luca, to determine the maximum loads that they could take under the given loading conditions. These results would, then, allow our MQP group to determine the necessary size of the femoral flap, with a set length of 10 cm, that was needed in order to surpass the, previously determined, fibula flap strengths. However, once experimental compression tests were conducted and completed for the four whole femurs, the results revealed further inaccuracies in the FEM mesh and, due to time constraints, our methodology had to be adjusted accordingly. In addition to the difficulties with the whole femur's mesh, there were complications in simulating 3-point bending of the femoral and fibula flaps. The attachments used to hold the flaps were pushing through their meshes because the program was not recognizing the meshes as solid surfaces. Luca worked on perfecting the mesh, in particular the mesh of the femur. Our MQP group developed other methods, as written below, to experimentally determine the appropriate width for the 10 cm

femoral wedge piece needed to surpass the strength of the 10cm cylindrical fibula flap.

Different sized graft pieces, from the femur, were tested under 3-point bending and compared to the fibula flaps. Femur flaps with a length of 10cm and circumferential percentages of 15%, 20%, 25%, and 30% (θ as seen in figure 5.2) were used to determine the wedge size needed to exceed the strength of the fibula flap from 5.1.1. Three 15%, three 20%, three 25% and one 30% femoral graft pieces were removed from the same cadaver in the region of the proposed femoral flap. Refer to Appendices J and K for the list and description of the bone specimens used. The circumference of each bone was measured about the middle of its shaft. The circumference measured was then multiplied by the appropriate percentage to determine the size of the segment that was to be removed. The 10cm long graft piece was measured in the middle of the shaft along the axis of the bone at the anterior distal lateral region of the vastus intermedialis and its width, which was determined in the previous calculation, was measured about the circumference. The graft piece was removed using an oscillating orthopedic saw and the bones were sprayed with saline during incision in order to keep the bones wet and to prevent the saw from burning the edges of the bone. The edges of the harvest site were rounded off using a Dremel Kit[®], once again spraying saline on the bone during the rounding process in order to prevent the Dremel[®] from burning the bone, in order to eliminate the stress concentration points at the top and bottom of the flap removal site. Upon removal of the graft pieces, the femur flaps and the femur's themselves were wrapped in saline-soaked gauze, and the femur's were put back in the fridge at 4° C, while waiting to be tested. Three point bending was applied and the strength of each flap size was compared to the results for the fibula flaps from 5.1.1 in order to determine the wedge size that was used in part 5.1.3.

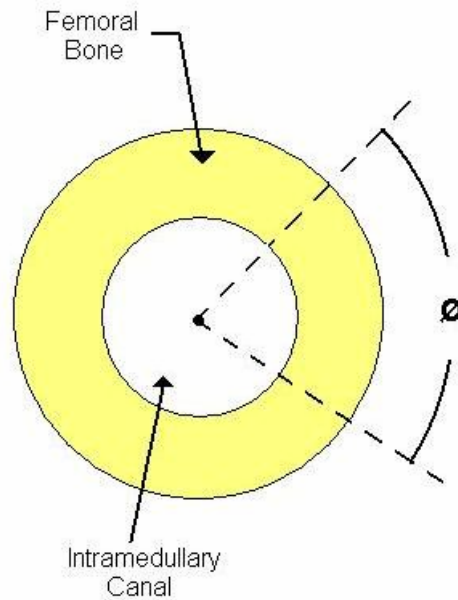


Fig. 5.2: This view is illustrates the cross-section of a femur and the theta is the angle of the circumference.

5.1.3 From the results in 5.1.2, five femoral grafts at 35% (θ) and four femoral grafts at 40% (θ), with a length of 10cm, were removed from the femurs. Refer to Appendices J and K for the list and description of the bone specimens used. The graft pieces were removed, stored, and tested according to the procedures described in 5.1.2. The 3-point bending was applied in order to compare the structural strength of these grafts. The femur grafts were loaded until failure, where the bone cracked, in order to determine the strengths of the different flaps. These results for the femur wedges were compared to the results from 5.1.1. This allowed for the strengths of the femoral graft pieces to be compared to the strengths of the fibula graft pieces in 3-point bending (Figure 5.3).

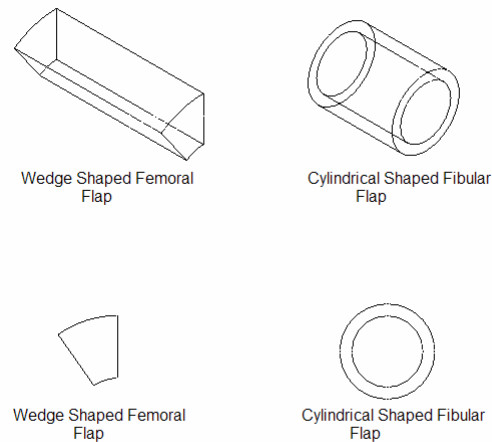


Fig. 5.3: Flap Geometries - for femoral and fibular flaps that will be structurally analyzed using three-point bending.

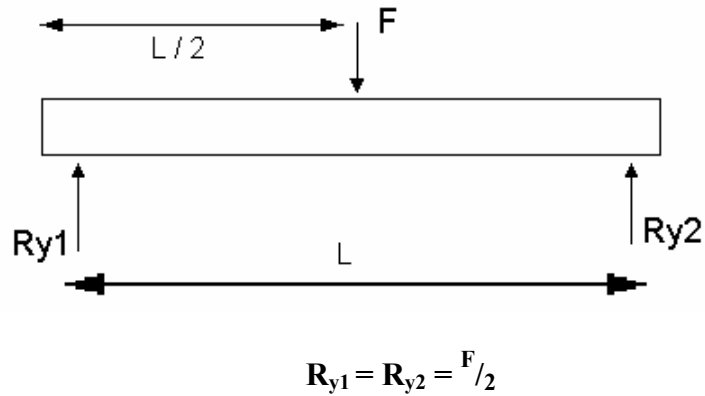
5.1.4 All data were checked for outliers using SPSS 12.0 for Windows. Q-Q plots were run and outliers were determined and eliminated. Statistical analysis of the data, without the outliers, was run using SigmaPlot 9.0 in order to test the trend seen in the data. Two-way ANOVA was used to compare the 35% femur flaps to the fibula flaps and the 40% femur flaps to the fibula flaps. Two-way ANOVA was run in order to apply blocking to the results, such that the differences among the specimens and the flap types (35% femur/40% femur or fibula) were taken into account by grouping them together because these factors affected the results. Refer to Appendix H for analysis procedure.

Analysis of Test Results

Stress- Strain

The three-point bending tests for the fibula and femur flaps collected load and displacement data. The displacement data was the distance that the crosshead had traveled downward during the test. In order to find the effective moduli and other structural properties, this data was used to calculate the stress and strain properties of the bone specimen. The structures were simplified by assuming that the materials were homogenous and linear elastic in behavior. This assumption allowed for the reaction forces created by the support abutment distribute the load of the crosshead equally. This led to the calculation of the internal moment

that the bone experienced point where the crosshead descended ($L/2$). A free-body diagram was used to illustrate all the forces involved (refer to Figure 5.4).



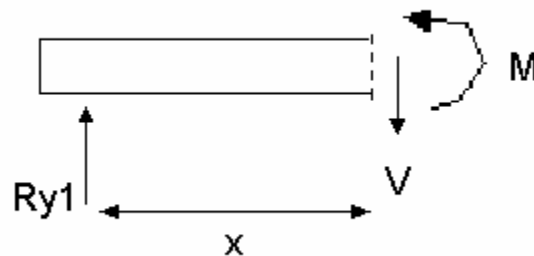
L = Length of specimen (0.1 m)

F = Force applies by the descending crosshead

R_{y1}, R_{y2} = Reaction forces where that bottom abutment supported the bone (10% and 90% of length)

Figure 5.4: Free-Body Diagram of 3-Point Bending

A cut was made at the point where the crosshead descends, to calculate the magnitude of the moment at that point (refer to Figure 5.5).



M = Internal moment experienced by the bone

V = Shear force experienced by the bone

x = Distance from R_{y1} to the cut (0.04m)

Figure 5.5: Internal Moment

This method allowed for the summation of all the forces and the moment was calculated using Equation 1. The internal moment increased during the test as the force measured by the load cell increased.

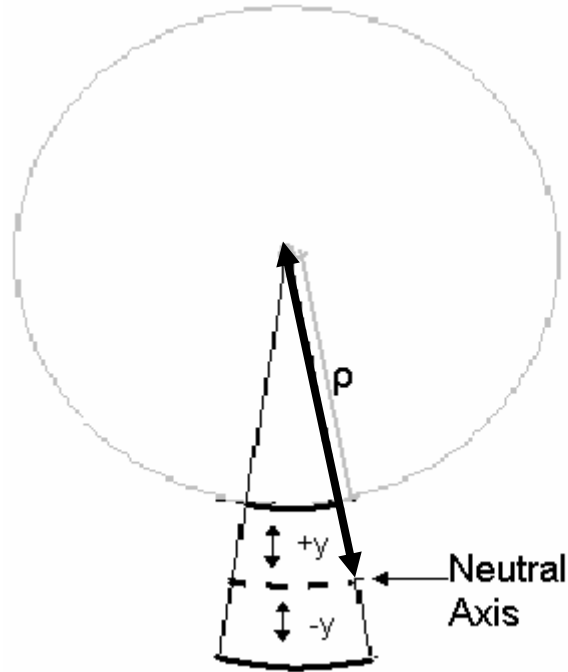
(Equation 1)
$$M = \frac{1}{2} F \cdot x \quad (\text{Hibbeler, 2000})$$

Using the *Flexure Formula* (Equation 2), the internal moment previously determined was implicated to find the stress (σ).

(Equation 2)
$$\sigma = - \frac{M \cdot y}{I} \quad (\text{Hibbeler, 2000})$$

In this equation, y is the distance from the neutral axis. The neutral axis is found at the centroid of the cross-section and does not experience stress. If the stresses at the top surface were sought, y would have been positive and vice versa if the stresses at the bottom surface is sought. In this study, the bone's weaker properties are in tension, which would be located near the bottom surface. The calculation of the centroid and the area moment of inertia, I , is later discussed.

To determine the strain, the material was again assumed to be homogenous and linear elastic. The data provided the displacement of the crosshead as it descended downward. As it descended, the bone experienced a deflection, which can be measured using the *Radius of Curvature* (ρ). The radius of curvature was measured by super-imposing a circle to fit the arc that was created by the deflected specimen. As the deflection increased, the size of the circle and its radius decreased (refer to Figure 5.6).



ρ = radius of curvature
 $+y$ = distance from neutral axis upward
 $-y$ = distance from neutral axis downward

Figure 5.6: Radius of Curvature

The circle was geometrically analyzed to find the magnitude of the radius. The contact points of the abutments were used as reference points to determine x and y and so the radius was calculated using Equation 3.

(Equation 3)
$$\rho = [1+(y')^2]^{3/2} / \text{abs}(y'')$$
 (Varberg, 2000)

The radius of curvature was then implicated in Equation 4 where strain was equal to the change in length per original length percentage ($[x-x_0] / x$).

(Equation 4)
$$1/\rho = -\epsilon/y$$
 (Hibbeler, 2000)

The y is the distance from the neutral axis where no strain is experienced by the bone specimen.

Properties of Area of Flaps

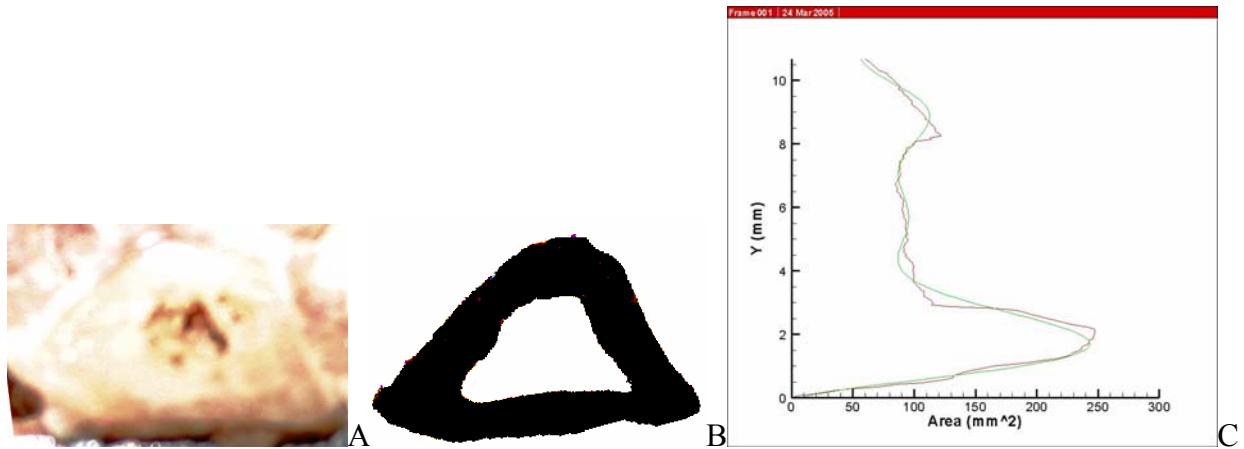


Fig 5.7: Photo Analysis A) Original cross-section image of fibula flap, B) cortical bone is made threshold, C) the change in area as in relation to the change in the y-axis (notice the smoother red polynomial fit).

The data recorded from the 3-Point Bending Tests of the Fibula and Femur Flaps were loads and displacements. The displacements referred to the distance that the crosshead descended during the time of loading. This data was used to determine the stress-strain information regarding the bone structures. To finish the calculation of the stress and strain properties, the area properties of the specimen's cross-section were needed. Due to the structural complexity of the bones, the images of sample cross-sections were analyzed using image analysis methods (refer to Figure 5.7). Post-experimentation images were taken of both ends of the fibula for this analysis with a metric ruler to establish pixels per millimeter calibration. Using Scion Image 1.63 for MacOS 7.5 to 9.x. Acquisition and Analysis Software (Scion Corporation, Maryland), the cortical areas of the cross sections were made threshold. This program also provides a *Plot Profile* option, which supplies a data listing for the change in area in relation to the y-axis. This data was uploaded into Tecplot version 10.0 release 3 (Tecplot Incorporated, Washington). This program was capable of fitting a 10th order polynomial curve ($F(y)$) to the data curve (Equation 5). Capitalized letters symbolizes the constants in this equation computed by the Tecplot program.

(Equation 5) $F(y) = A + By + Cy^2 + Dy^3 + Ey^4 + Fy^5 + Gy^6 + Hy^7 + Iy^8 + Jy^9 + Ky^{10}$

The program was used to upload this equation to MAPLE 7 (Waterloo Maple Incorporation, Ontario, CA). This math program was used to calculate the centroids of each

cross section as well as the Area Moments of Inertia about the x' axis. The x' axis is the location of the neutral axis, which is at the centroidal axis. The Area Moment of Inertia about the x' axis is described by the *Parallel Axis Theorem* (Equation 6 and Figure 5.8).

(Equation 6)
$$I_{x'} = \int_A (y' + d_y)^2 dA$$
 (Hibbeler, 2000)

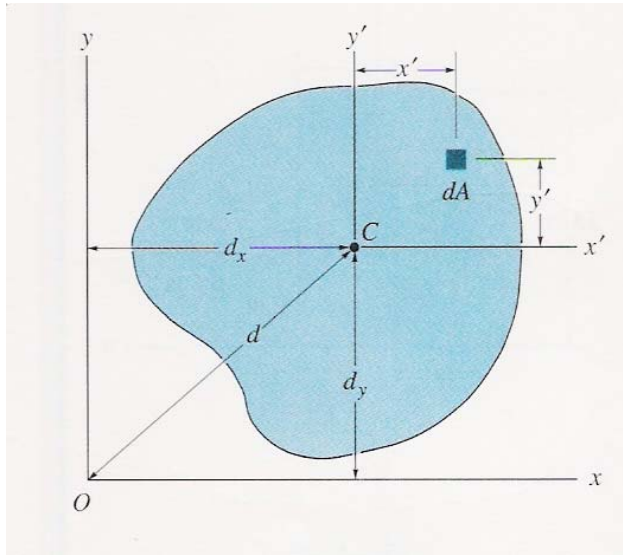


Fig 5.8: Parallel Axis Theorem (Hibbeler, 2000)

In this study, y' (the distance from the x' axis to the desired axis is zero). The distance from the bottom of the bone to the centroid is represented by the variable d_y and dA is the change in the area. This formula is implemented into the following program:

```

> a:=constant;
> b:=constant;
> c:=constant;
> d:=constant;
> e:=constant;
> f:=constant;
> g:=constant;
> h:=constant;
> i:=constant;
> j:=constant;
> k:=constant;
> m:=a+b*x+c*x^2+d*x^3+e*x^4+f*x^5+g*x^6+h*x^7+i*x^8+j*x^9+k*x^10;
> plot(m,x=0..10);
> r:= "maximum value of y";
> Area:=int(m,x=0..r);
> z:=int(m^2,x=0..r);
> C:=z/Area;
> Q:=int(C^2*m,x=(C-.5)..(C+.5));
> Inertia:=Q*1E-12;

```

This program carries out the integrations for each cross section to measure the Area Moment of Inertia (Inertia) Centroid (C). These calculations were done for each cross section and applied to the *Flexure Formula* (Equation 7).

(Equation 7)
$$\sigma = - M y / I$$

This equation was used because the material of the bone was assumed to be linear-elastic and homogenous. This assumption does not represent the bone as described in the background; bone is nonlinear elastic and is not homogenous. However, for simplicity, these assumptions were made. This equation is used to calculate the stress (σ) experienced by the material a y distance away from centroid. The stress was calculated on the bottom surface of the bone where tension is experienced. Using the calculated stresses and strains, graphs were plotted to determine the ultimate tensile strengths and effective moduli for each specimen and each structure on average. (The imaging procedure is described in depth in Appendix I)

Specific Aim II:

The second specific aim was to determine the structural integrity of the harvested femur. The harvested femur must not exceed its yield strain while being loaded under fiftieth percentile male body weight during stair descent (2447N) after the minimal circumferential percentage is removed.

5.2 Compression Testing

The femoral specimens and abutments were loaded by a hydraulic MTS loading machine Sintech 5/G ® (Highway and Infrastructure Laboratories, Civil Engineering Department, Worcester Polytechnic Institute). The bones were loaded in compression at a strain rate of 0.1 mm/sec, as to provide a loading that replicated natural loading during ambulation. This strain rate, also, allowed for precise reading of load-strain curves. The femur specimens were loaded to failure, where the bone fractured, and observations were made. These methods were adopted from Knothe et al.

5.2.1 Compressive Fixation

The condyle of the femur was fixed in a Free Tree Stand Abutment that fastened to the bone using cement potting techniques. Fine anchoring cement was mixed with water and poured into coffee cans, whose top halves were sawed off, in order

to anchor each of the whole and harvested femurs. The lower portion of the femur condyle was submerged in the cement and the femur was immediately aligned in the coffee can, using a protractor, before the cement started to set. The femur was orientated in the pot as it would be orientated in the body when an individual is standing, as seen in figure 5.9. A ring stand was used to hold the potted femur in the aligned position until the cement set. The bones were sprayed with saline during the cement drying process, avoiding the cement itself, in order to keep the bones wet. Upon the cement drying, the potted femurs were re-wrapped in saline-soaked gauze and put back in the fridge at 4° C while waiting to be tested.

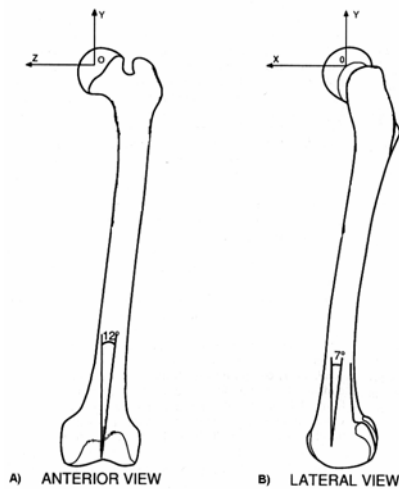


Fig. 5.9: Femur Orientation - A) Bicondylar angle of 12°, and B) 7° angle of adduction (Christofolini, 1995).

5.2.2. Alignment with Abutments

The potted femur was placed in the lower abutment and the femoral head was positioned in the upper abutment, which had a concave region that limited the horizontal motion but allowed for the rotation of the femoral head. This is seen in figure 5.10. The threaded screws were tightened against the bottom abutment, to secure it in place, once the femur was aligned with the upper abutment. Refer to Appendix F for the design of the attachments.

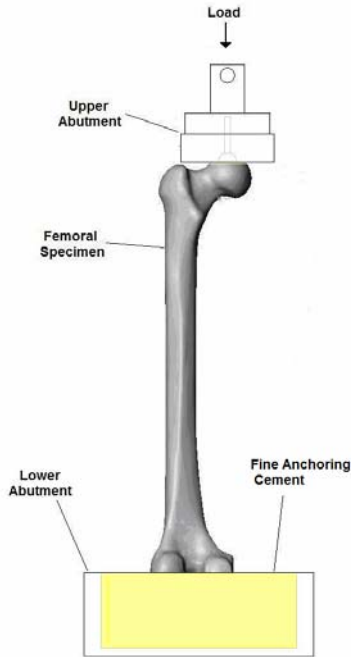


Fig. 5.10: An illustration of the compressive loading on the femoral specimen.

5.2.3 Strain Gauges

Strain Gauges were positioned around the donor site to measure the strains during loading. It was assumed that, due to the locations of the strain gauges and the orientation of the femur, strain gauge 2, as seen in figure 5.11, would most likely experience compression during loading while the rest of the gauges would experience tension. By orientating the gauges around the donor site, the highest strain due to tension was acquired. The positioning of the gauges is illustrated in Figure 5.11. The strain gauges were applied to the bone by placing a thin-sanded layer of CPC onto the gauge site, using UV light to cure it, and then attaching the strain gauge to the CPC using super glue. The strain gauge wires were attached to the computer through a converter with the help of Vincenzo Bertoli, a civil engineering graduate student.

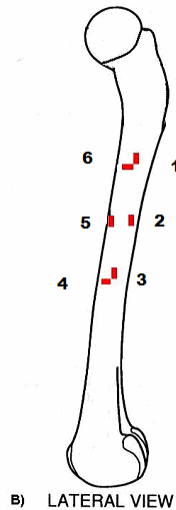


Fig. 5.11: Positioning of Strain Gauges around the donor site.

- 5.2.4 The whole femur bones were loaded in compression and tested to failure with strain gauges surrounding the site where the segment was removed. Four whole femurs were used in testing. Data from these tests, that was later used to construct the FEA simulation, came from the strain gages that were placed on the femurs.
- 5.2.5 Originally, after the femoral wedge size was determined by Luca using FEM, as mentioned in 5.1.2, the femur structure, with the removed graft piece, was going to be simulated in anatomical position under uniaxial compressive loads. This was going to be used to determine if the harvested femur would need to be rodded in order to prevent it from passing its yield strain. However, the difficulties with the FEM, mentioned in 5.1.2, revealed that the experimental methods had to be adjusted. As a result, the methods below were followed.

The femurs with the previously removed 35% and 40% graft pieces were loaded in compression and tested to failure, where fracture occurred. Five 35% harvested femurs and four 40% harvested femurs were used in testing. Refer to Appendices J and K for the list and description of the bone specimens used. Data from these tests, that was later used to construct the FEA simulation, came from the strain gages that were placed on the femurs.

5.2.6 All data were checked for outliers using SPSS 12.0 for Windows. Q-Q plots were run and outliers were determined and eliminated. Statistical analysis of the data, without the outliers, was run using SigmaPlot 9.0 in order to test the trend seen in the data. T-tests were used to compare the 35% harvested femurs to the whole femurs and the 40% harvested femurs to the whole femurs. The one tailed t-tests were used to statically determine if the data from the 35% harvested femurs or the 40% harvested femurs was significantly different from the data for the whole femurs. This, in turn, revealed whether or not the femoral structure was significantly affected by the removal of the graft piece. Refer to Appendix H for analysis procedure.

Analysis of Test Results

The mechanical testing procedures provide force – strain curves. These curves are later used to provide the ultimate force the specimen can withstand, as it relates to the size of the osteotomy.

This data shows how the ultimate force of the structure decrease as the size of the osteotomy increases. The ultimate forces should always be greater than the desired 550 lbs load. If the ultimate force comes to be smaller than this desired load, the data shows that the bone is incapable of supporting the weight of a 50th percentile male as he is descending down stairs. This data has been used to initially determine if θ , which is the size of the wedge that must be removed from the femur in order to surpass the strength of the fibula, weakens the structure excessively. Similarly, the stress-strain curves are used to determine the Ultimate Stress, which is depicted as the highest point on the curve prior to a rapid descent and catastrophic failure.

The strain gauges used during the mechanical testing provided information to construct the Finite Element Model. Luca Valle, in the civil engineering department, used this data to create and test these models in FEA. Through this FEA tests, the force at which the structure yields at was determined. Furthermore, the strain gauges provided stress readings around the donor site as to better understand the structure of the femur and how the osteotomy impacts the structural integrity at a local level.

5.3 Using Materials Testing to Construct Finite Element Models (FEM)

We worked in collaboration with a group of graduate students in the civil engineering department at Worcester Polytechnic Institute. We worked together in order to validate their Finite Element Model of the fibula flap and of the femur, as well as to determine whether or not the harvested femur yielded prior to reaching the force occurring in a 50th percentile male's femur in stair descent. This force is the maximum static force loaded on the femur. Luca Valle, who worked most closely with us, developed a model of the femur in FEM using an existing model from BEL Repository (G. Cheung, et al, 2004). However, there were errors in the acquired mesh of the model. Luca used data from our team's background research, such as the material properties of cortical and cancellous bone, in order to create an accurate mesh for the geometries and properties of the femur at the condyle, at the head, and at the neck. He, also, added a cancellous layer of bone inside the cortical bone. Additionally, we provided Luca with the material properties of the fibula and he modeled a fibula segment in the shape of a cylinder.

- 5.3.1 The mechanical testing results were used to construct computer simulated tests of the femur in LS-DYNA. This data was used by Luca Valle to construct the models of the femur, the femur flap, and the fibula flap. The abutments were modeled and incorporated, by Luca Valle, in FEA simulation of the tests to replicate the loading that was applied during the mechanical testing procedures.
- 5.3.2 The whole femurs and 40% harvested femurs were created and tested to failure, where fracture occurred, in FEA by Luca Valle. Since the strength of the 35% femoral flap matched, and did not surpass, that of the fibula flap, it was not necessary to conduct further analysis on the 35% harvested femur's structure using FEM (this test was ran regardless). The FEA loads at failure and the location of failure were compared to the mechanical test data to validate the model.
- 5.3.3 Once the FEM model of the femur was completed, with the appropriate mesh, Luca was able to run the test to determine if the harvested femur, loaded under uniaxial compression in the anatomical position, yielded prior to reaching the

force occurring in a 50th percentile male's femur in stair descent (2447N). This was achieved by determining the force at which the harvested femur yielded at in FEM, and then comparing it to the force on the femur in stair descent.

Bone Quantities Used

Due to the limited supply of bones provided by University of Massachusetts Medical School, the bone supply is distributed among all the tests proposed in this methodology.

Test ▼	Bone (total available) ►	Femur (20)	Fibula (20)
5.1.1			13 bones
5.1.2		3 bones	
5.1.3		9 flaps	
5.2.1		4 bones	
5.2.2		9 bones	

*5.1.2 uses several flaps from two bones. 5.2.2 uses the bones with segments removed in 5.1.3.

** Refer to Appendix J for a list of which specimen (cadavers) were used in each test.

***Refer to Appendix K for the description of the cadavers used in testing.

Chapter 6: Data

In this section the data from the procedures followed in the methodology are given. The data set that is given first is the three point test data of the fibula flap that would be used to reconstruct a non-continuous tibia defect of ten centimeters. The following set of data is from the initial regional studies of the femur harvest region, through three-point bending, to determine the approximate testing parentages. Next the data set is from the three point bending studies of the 35% and 40% circumferential femur flaps. The data set that subsequently follows comes from the compression testing of whole femurs. Finally that last data set given comes from the compression of host femur after the femoral flap has been removed.

Three Point Testing

Fibula Three Point Bending Testing

Data was collect from the three point bending test to allow for a comparison between the structure of a fibula flap and different femur flaps. Data from individuals that produced both fibula flaps and femur flaps was gathered to study the difference in structural properties of the flaps within the same individual. This data was gathered in a force verse displacement graph, such a graph is shown below for individual 1975 (Figure 6.1). The complete data set can be found in Appendix L.

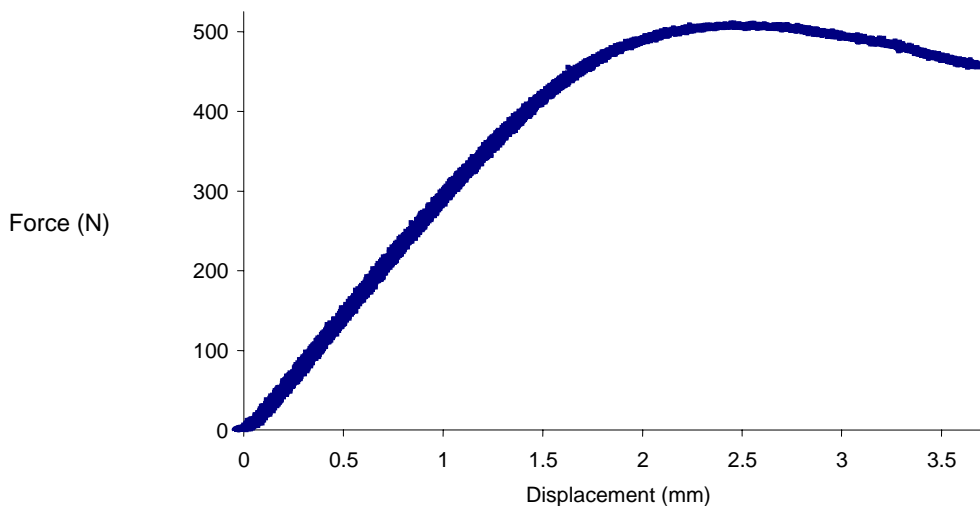


Figure 6.1: Force versus Displacement Curve of a Fibula Flap in 3-Point Bending for individual 1975a

The data collected from these graphs is represented in the table below (Table 6.1)

Table 6.1: Resultant Maximum Loads and Yielding Loads of All Fibula Flaps

<u>Fibula Three Point Bending Results</u>		
Individual	Max Force (N)	Yielding Force (N)
1975 a	511	445
1975 b	383	284
2000 a	612	587
2000 b	705	510
2008 a	759	620
2008 b	652	548
2017 a	564	559
2017 b	487	474
2020 b	1120	1050
2028 a	497	463
2028 b	513	491
2031 a	566	483
2031 b	767	589
AVERAGE	626 ±186	546 ±174

Initial Regional Studies of the Femur Harvest Region

The regional studies of the femur harvest region allows for an evaluation of the bone structural properties. This test allowed for a circumferential percent to be determined for the bone flap. Multiple flap percentages were experimented on in three point bending tests. The data was collect within two individuals with multiple flaps harvested form the region. The data was collect in force-displacement curves; a curve for this data type is given below in Figure 6.2. The complete data set can be found in Appendix M.

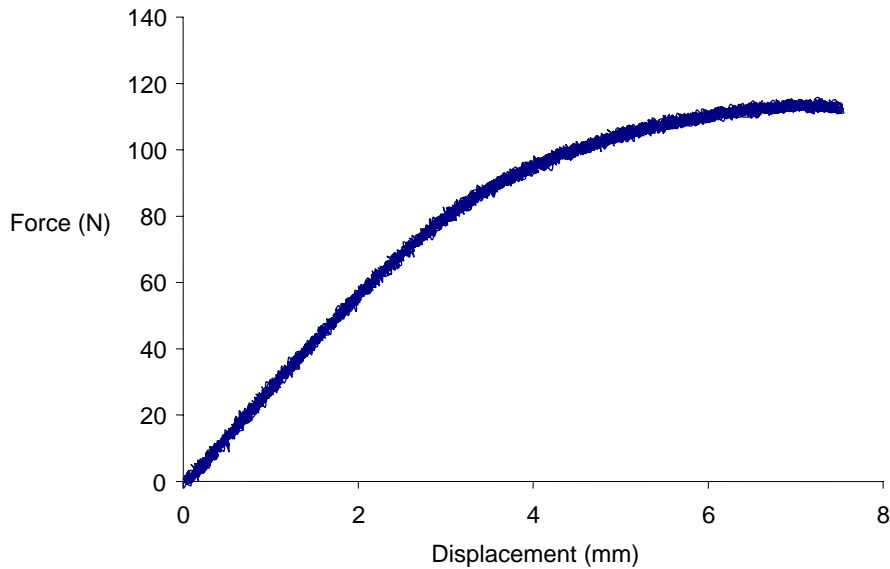


Figure 6.2 Force Versus Displacement Curve of Regional Studies 15% Femur Flap for individual 2008

This force-displacement curve represents the individual 2008, and the percent circumference tested is 15%. The data collected from these curves is represented in the table below (Table 6.2)

Table 6.2: Resultant Maximum Loads When Determining Circumferential Percentage

Initial Regional Studies Compression Results				
Individual	Cir. %	Max Force (N)	Max Displacement (mm)	
2008	15	194	7.4	
2008	15	113	7.55	
2008	15	77.3	9.16	
2008	20	103	8.54	
2008	20	227	4.77	
2028	20	82	7.57	
2008	25	304	5.51	
2028	25	149	8.03	
2028	25	129	6.42	
2008	30	622	4.02	
		AVERAGE 15%		
		128 ±59.8		
		AVERAGE 20%		
		137 ±78.4		
		AVERAGE 25%		
		194 ±95.8		

Studies of the 35% and 40% Circumferential Femur Flaps

The studies of the 35% and 40% circumferential femur flaps in three point bending allows for an evaluation for the strength of these two structure compared to a fibula flap. The data was collect in force-displacement curves; curves for 35%, 40% and 35% & 40 % (for the same individual) is given below in figures 6.3, 6.4, 6.5. The complete data set can be found in Appendix M.

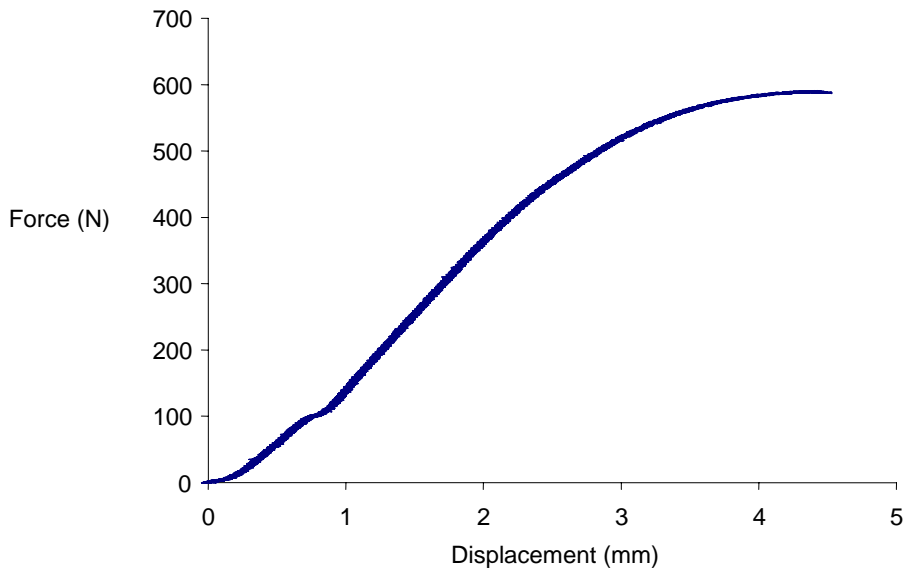


Figure 6.3: Force Vs. Displacement of 35% Femur Flap-shows the force needed to displacement individual's (1975) femur flap (at 35%) in three point bending.

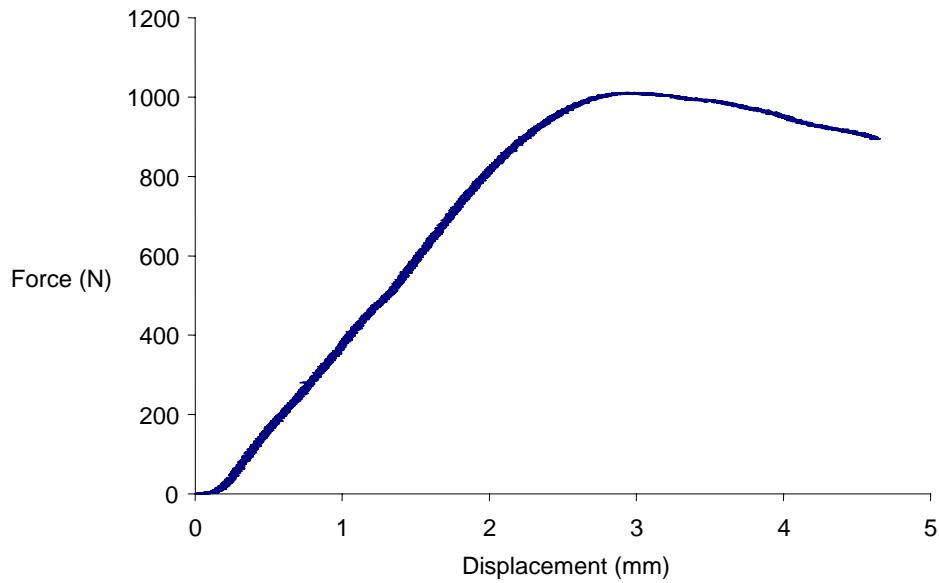


Figure 6.4: Force Verses Displacement of 40% Femur Flap shows the force needed to displacement individual's (1975) femur flap (at 40%) in three point bending.

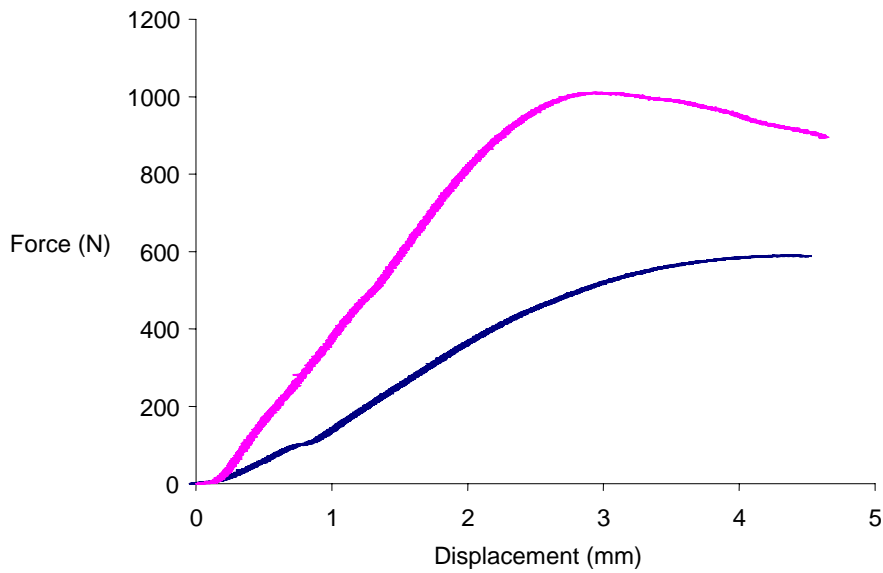


Figure 6.5: Force Verses Displacement (35% Vs. 40%) - shows the force needed to displacement two different sized femur flaps (35% (blue) and 40% (purple)) from the same individual (1975).

The graphs are summarized in table 6.3 and figure 6.6 provided below.

Table 6.3: Resultant Maximum Loads of 35 and 40 Percent Femoral Flaps

Femur Flap Three Point Bending Results				
Individual	Cir. %	Max Force (N)	Max Displacement (mm)	
2000	35	948	4.7	
2017	35	771	4	
1975	35	590	4.53	
1975	40	1010	4.66	
2020	35	1240	4.9	
2020	40	1940	3.56	
2031	35	797	6.05	
2031	40	1510	5.64	
2028	40	439	5.78	

AVERAGE 35%
869 ±243

AVERAGE 40%
1220 ±647

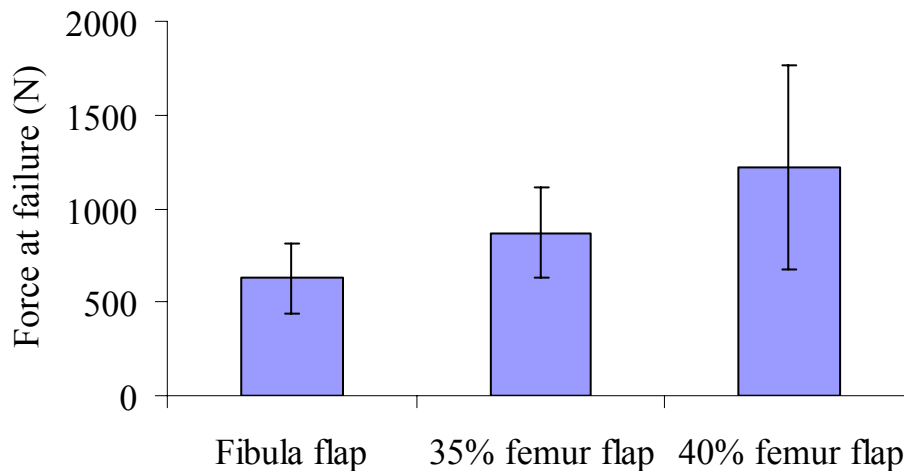


Figure 6.6-Femur Verses Fibula Flaps in Three Point Bending. n=13 (Fibulas), n=5 (35% flaps), n=4 (40% flaps)

Compression Testing

Whole Femurs

Data was collected for the structural properties of the whole (non-harvested) femur to allow for an understanding of the effect of removing the flap from the femur. The whole femurs were placed in anatomical orientation during uniaxial compression. A graph of this data is given below in Figure 6.7.

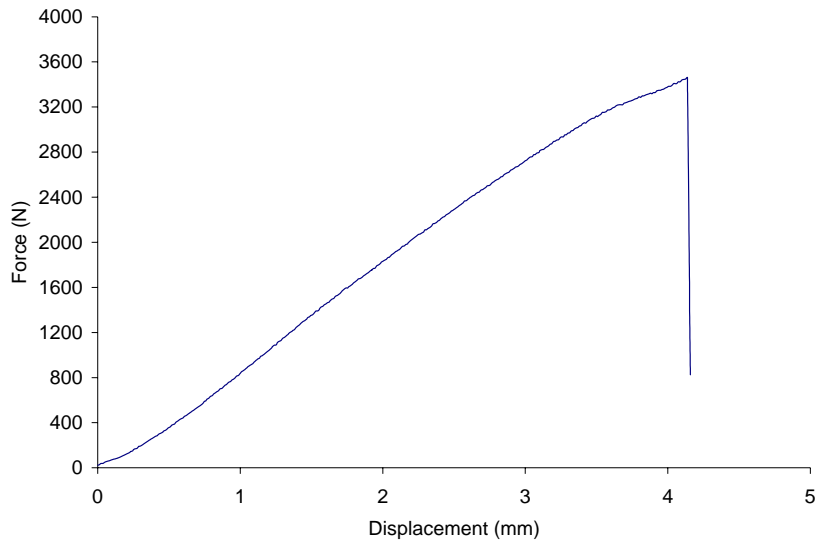


Figure 6.7: Full Femur Compression - represents the full femur of individual 2017 (right side).

The rest of the femurs overall displacement, and the forces to cause such a displacement, are all given in Appendix N; and summarized on 6.4.

Table 6.4: Resultant Maximum Loads in Whole Femurs
Full Femurs Compression Results

Individual	Max Force (lbs)	Max Force (N)	Max Displacement (inches)
2000 R	528	2350	0.206
2009 L	1090	4850	0.326
2009 R	1180	5250	0.313
2017 R	778	3460	0.261
AVERAGE	894 ±299	3980 ± 1330	0.277 ± .0548

Femurs After Removal of Flap

Data was collected for the structural properties of the harvested femur to determine the effects of removing the flap from the femur. Femurs with 35% and 40% circumference sized flap removed were tested in anatomical position. Below are the results of the compression test of individual 2017 that had a 35% circumference flap removed (Figure 6.8). The complete data set can be found in Appendix N; and is summarized in table 6.5 and figure 6.9.

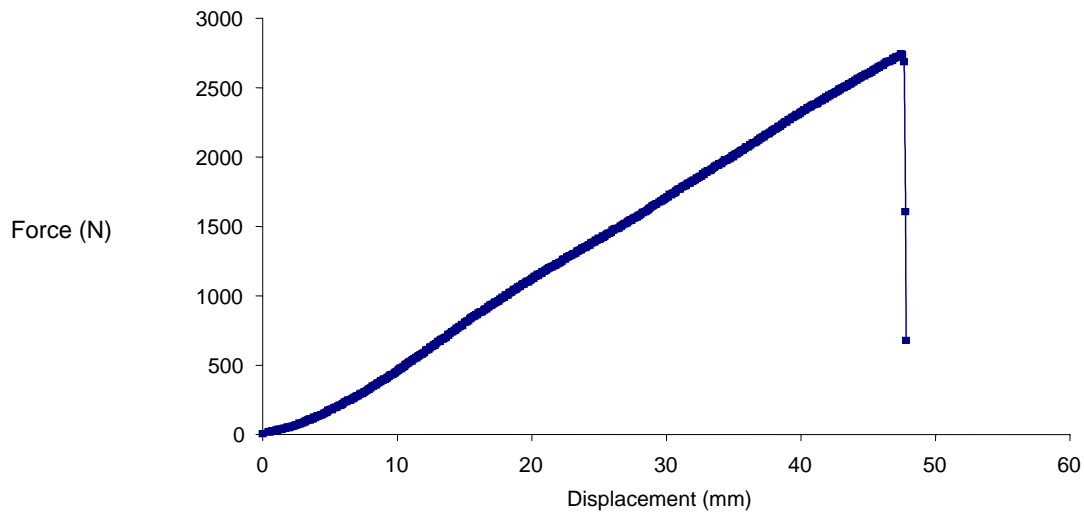


Figure 6.8: Force-Displacement Curve of a Femur with a 35% Flap Removed for individual 2017

Table 6.5: Resultant Maximum Loads in Femurs with 35% or 40% Flaps Removed

Harvested Femurs Compression Results				
Individual	Cir. %	Max Force (N)	Max Displacement (mm)	
2000 L	35	2860	44.9	
2017 L	35	2740	47.5	
1975 L	35	4340	45.1	
1975 R	40	4182	51	
2020 L	35	5080	60.9	
2020 R	40	5270	40.4	
2031 L	35	3000	48.2	
2031 R	40	3270	34.7	
2028 L	40	1250	54.7	
	AVERAGE 35%	AVERAGE 40%		
	3600 ±1050	2790 ±1700		

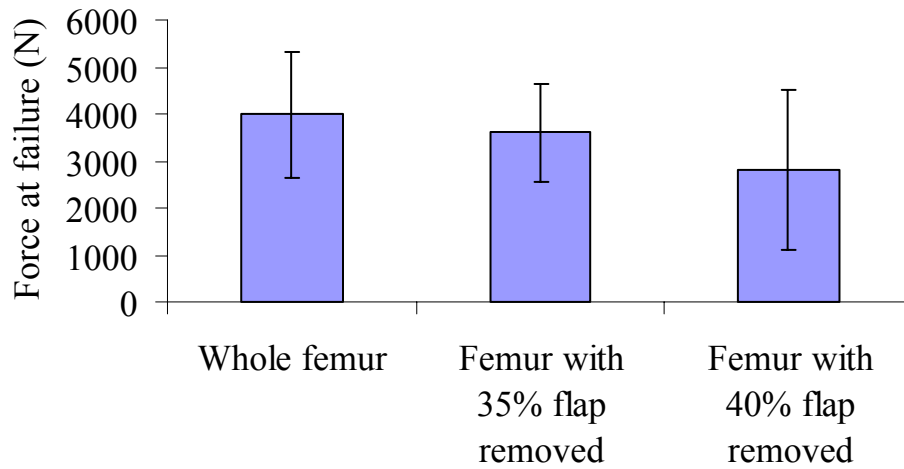


Figure 6.9-Compression Study of the Femurs. n=4 (whole), n=5 (35% removed) , n=4 (40% removed)

Finite Element Analysis

The test results of the femurs were placed into a finite element model. This model of the femur ran parallel test to the test ran on the real femurs. The data for the uniaxial compression test of a full femur, a 35% harvested femur, and a 40% harvester femur are given below on figure 6.10.

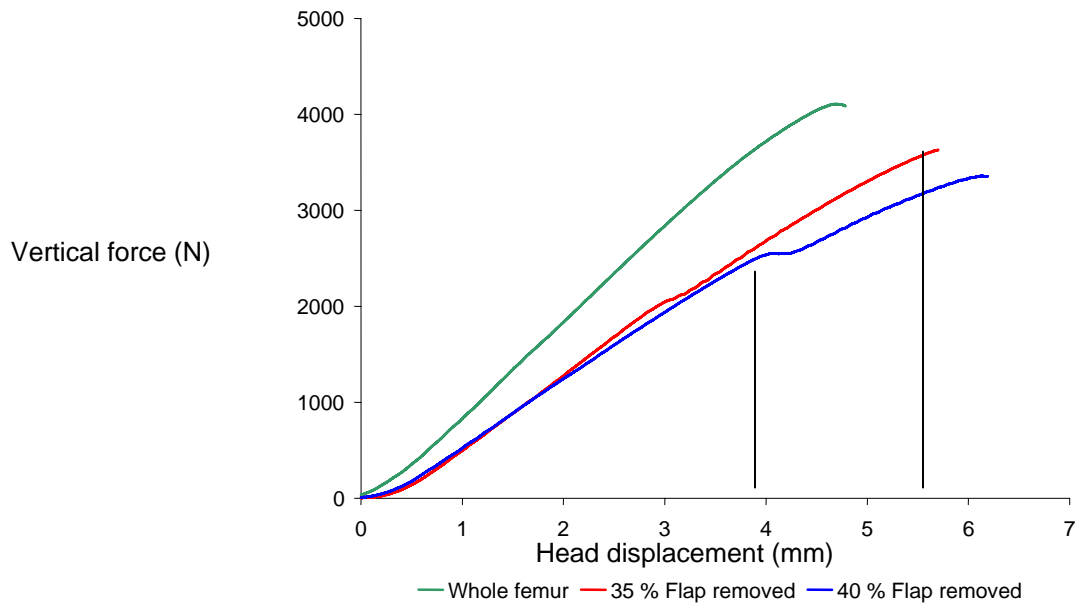


Figure 6.10: Force-Displacement Curve from FEA - of a whole femur, a femur with a 35% flap removed, a femur with a 40% flap removed

The head displacement when the first point in the 35% harvested femur had 1.8% strain, moving from elastic to plastic deformation, was at 5.616 mm. The force that corresponds to this displacement is 3604N.

The head displacement when the first point in the 40% harvested femur had 1.8% strain, moving from elastic to plastic deformation, was at 3.798mm. The force that corresponds to this displacement is 2443N.

Given in figure 6.11 and figure 6.12 are the results of the finite element analysis of the harvested femur for stress concentration prior to failure and failure.

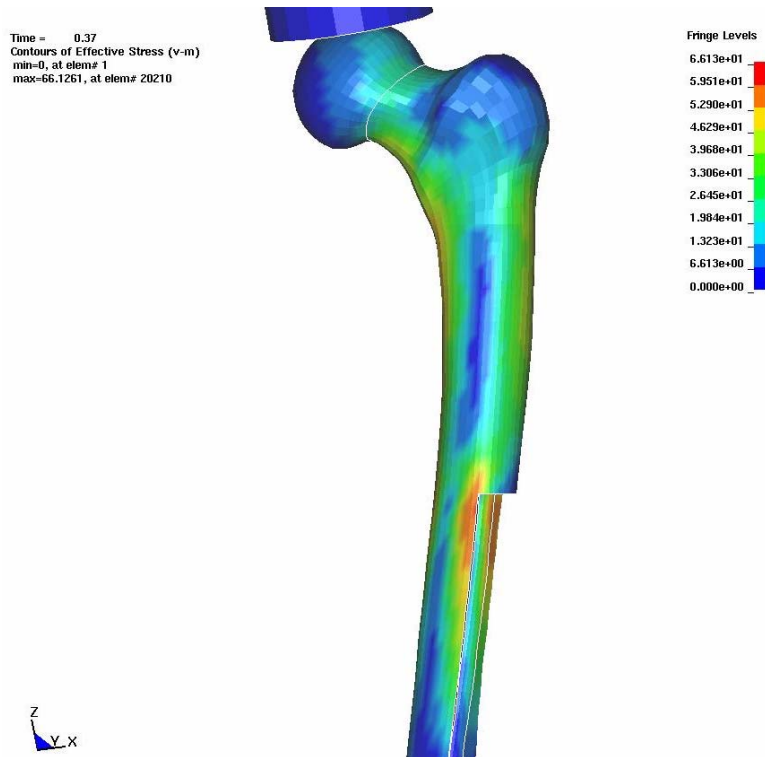


Figure 6.11 Femur Model Prior to Failure- A stress concentration (Red) at the proximal cut from where the flap was removed.

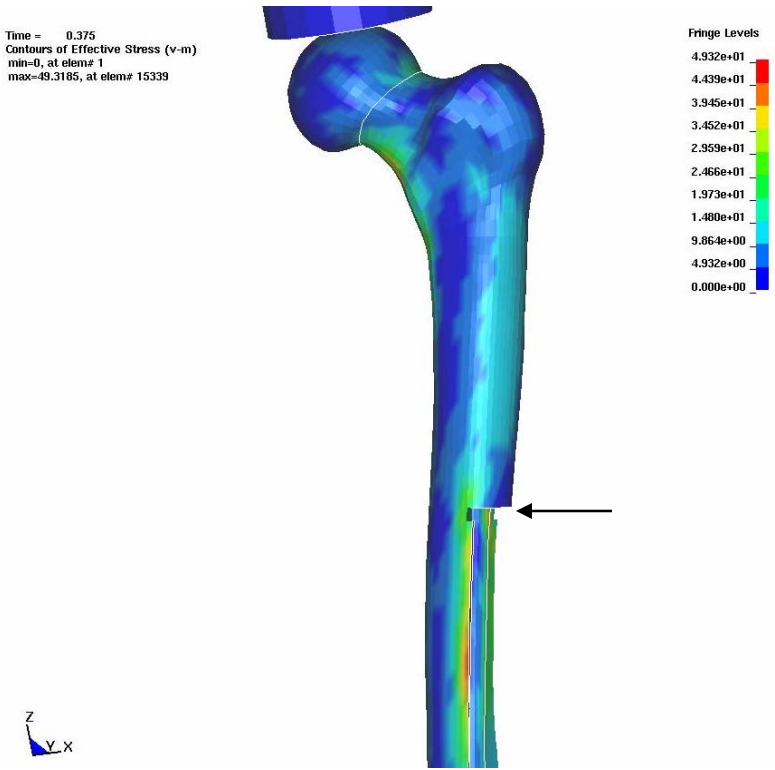


Figure 6.12 **Femur Model at Failure**-Deletion of elements signifies fracture failure.

Chapter 7: Analysis and Discussion

Specific aim one was met through the comparison of femoral segments and fibular flaps in 3-point bending experimentations. The results showed that the 35% femoral segment matches the maximum load of the fibular flap. However, statistical results show that the 35% femoral segment does not exceed the fibular maximum load by 100N. On the contrary, the 40% femoral segment exceeds the maximum load of the fibular flap by 225N ($p = 0.047$). This result shows that the femoral flap is capable of tolerating a higher maximum load and there for less likely to fail under post-operative mechanical stress as compared to the fibular flap. Because no previous evaluations have been completed of the newly developed femoral segment, there is a lack of data to compare experimental results.

Statistical tests were run using Sigma Plot 9.0 in order to compare the results from the 3-point bending tests. Two-way analysis was used to compare the flap results in order to account for the difference in specimens. The results were blocked according to specimen and flap type, fibula or femur, and two-way ANOVA was run for comparison of the 35% femur flaps and the fibula flaps, as well as the 40% femur flaps and the fibula flaps. The first test was run to test the hypothesis that the maximum force that the 35% femur flap can take in 3-point bending is greater than the maximum force that the fibula flap can take in 3-point bending. The maximum force that the fibula flaps can take in 3-point bending is 626 ± 186 N and the maximum force of the 35% femur flaps is 869 ± 243 N. A 95% confidence interval, where a significant P-value is less than 0.05, shows that the 35% femur flaps and the fibula flaps are significantly different ($P=0.002$, $n=13$ fibulas, $n=5$ 35% femur flaps). Thus, the 35% femur flaps are proven to be stronger than the fibula flaps. However, in order to account for a factor of safety, the femur flaps' maximum force in 3-point bending needs to be 100N greater than that of the fibulas', in order for the femur flap to be considered a suitable match to the fibula flap in tibial reconstruction. When taking this safety factor into account, it is found that the 35% femur flaps are not significantly different from the fibula flaps ($P=0.076$, where the confidence interval is set to a value of $P < 0.05$) and thus the 35% femur flap is not suitable for use in reconstruction of tibial fractures. The same tests were run in order to test that the 40% femur flap is stronger than the fibula flap in 3-point bending. The maximum bending force that the 40% femur flaps can take in 3-point bending is 1220 ± 647 N. The results show that the 40% femur flaps are significantly different from the

fibula flaps ($P= 0.003$, $n= 13$ fibulas, $n=4$ 40% femur flaps), where the factors are significantly different if P is less than 0.05. Therefore, the 40% femur flaps are stronger than the fibula flaps. When taking the 100N safety factor into account, it is found that the 40% femur flaps are significantly different from the fibula flaps ($P= 0.011$), in fact the 40% femur flap is 225N stronger than the fibula flap ($P= 0.047$). Therefore, it can be concluded that the 40% femur flap is, structurally, a suitable alternative to fibula flaps in reconstruction of tibial fractures.

The stress-strain curves plotted for the fibulas using Microsoft Excel (Microsoft Corporation) were used to evaluate the material properties of the specimen. The stresses showed to be 1000 MPa and the structures were observed to be quite brittle. The brittle property is implied by the small amount of strain required for the structure to fail. Refer to Figure 7.1 for average fibular results.

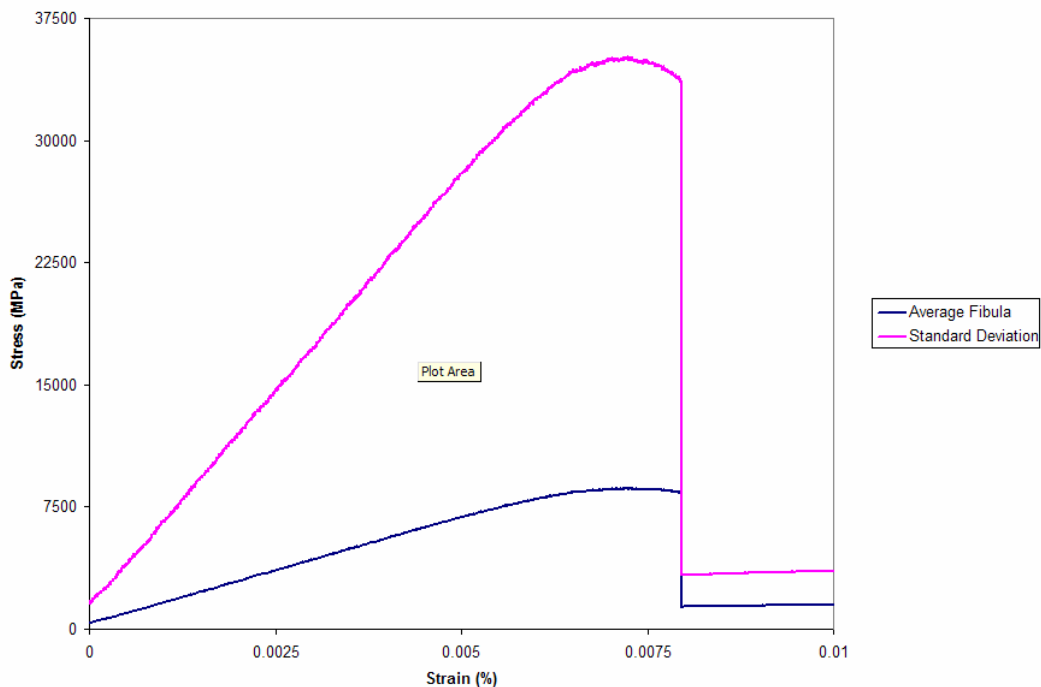


Fig. 7.1: Average Stress-Strain curve of Fibula Flaps with standard deviation

The femur flaps appear to be more brittle, as they take less strain to fail than the fibulas do. The 35% and 40% femur flaps were compared, and it was detected that the 40% femur flap was able to withstand a higher stress prior to failure as well as tolerating greater strain. The average femur flap stress-strain curve shows that the femur flap fails at a much higher stress than the fibula flap (refer to Figure 7.2). (See Appendix N for 35% Femur Flap data)

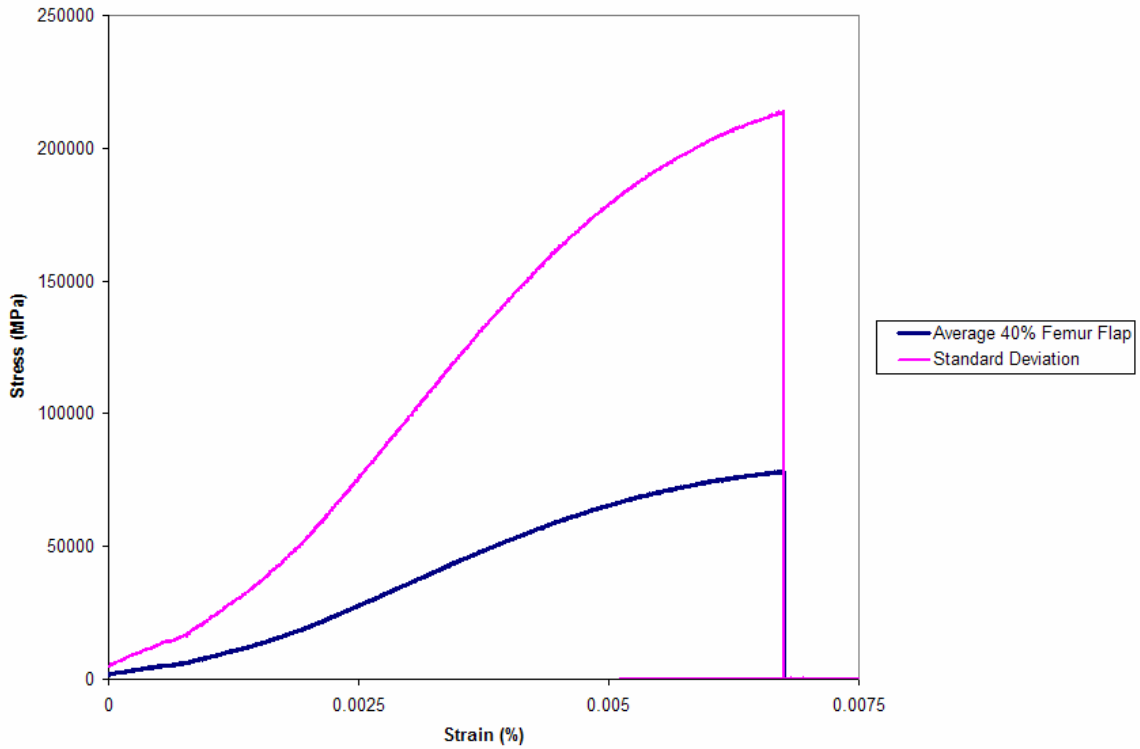


Fig. 7.2: Average Stress-Strain Curve of 40% Femur Flaps with standard deviation

The Comparison between the fibulas and femur flaps shows that on average the 40% femur flap structure tolerates a much greater stress than the fibular structure and the structure of the 35% femur flap. The data shows that the 35% femur flap fails at a lower stress and less strain than the fibular structure. These findings are illustrated in Figure 7.3.

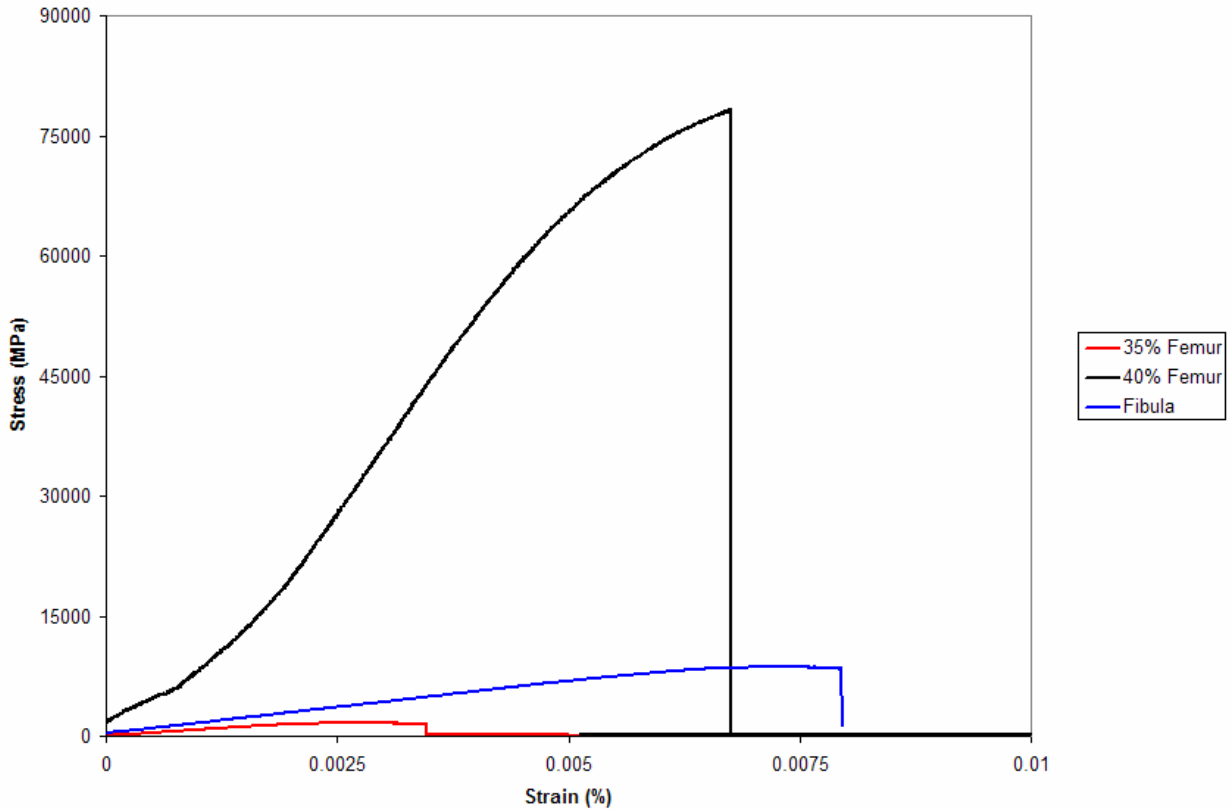


Figure 7.3: Average stress-strain curves for the 35% and 40% femur flaps in comparison to the fibular structure. The maximum loads of the 35% femur flap and the fibular flap more closely replicate the findings of Martin et al., where the Ultimate Tensile Strength in the longitudinal axis of cortical bone was 113 MPa (refer to Table 2.1). The Ultimate Tensile Strength may have increased as a result of embalment in 10% formalin. Literature reviews showed that there is a 16.3% increase in Elastic modulus, as well as a 10% increase Ultimate tensile strength. Furthermore, the strain percentage was said to decrease by 20.6%. All of these results, however, were found to be statistically insignificant (Sedlin, 1965; Evans, 1973). On the contrary, the stress-strain results suggested that all of these effects may be true as both the ultimate tensile strength and the effective moduli of each structure are drastically larger and the strain percentages are lower than the recorded material properties of human bone in Table 2.1. The results showed that the strain decreases from a range of 2.9% - 3.2% to 0.35% - 0.8% (~80% decrease) and the ultimate tensile strength increases from 133 MPa to about 29,000 MPa (~220% increase). The tensile elastic modulus increased from 9,600 MPa to $\sim 59.0 \times 10^5$ MPa (fibula $\sim 12.9 \times 10^5$ MPa, 40% $\sim 15.8 \times 10^6$ MPa, and 35% $\sim 7 \times 10^5$ MPa) showing an increase of about 600%. These numbers may be

skewed since the specimens are elderly individuals. However, the patterns follow similarly to those of previous findings.

Furthermore, Yamada's studies of the tibial, femoral, and fibular diaphysis in 3-point bending were slightly appeared in the results of our experiments. The 40% femoral flap was observed to have a higher effective modulus than the fibula, as illustrated in Yamada's finding in the 1970's (refer to Figure 2.2). Also in support of Yamada's findings, both femoral flaps failed at a smaller strain percentage than the fibular flap. The 35% flap fails at a lesser ultimate tensile strength than the fibula; however, this can be attributed to the small wedge-like structure of the 35% femoral flap as opposed to the cylindrical specimen used by Yamada. (Yamada, 1970). Refer to Statistical Analysis Results in the Appendix H.

Specific aim two was met through the use of an FEA model that simulated the average experimental results of the harvested femurs under uniaxial compression. The results showed that the 35% harvested femur yielded at 3604N, which is greater than the force that the femur will be subjected to in a fiftieth percentile male in stair descent (2447N). This means that the femur will not exceed its elastic limit when being loaded by a fiftieth percentile male trying to descend down a flight of stairs. However, because this is not a superior substitute to the fibula flap, the effects of the 40% segment removal were analyzed. Results showed that the 40% harvested femur yielded at a force lower than 2447N (2443N). This data indicated that the 40% harvested femur is structurally compromised and therefore requires restructuring. Previous studies using uniaxial compression in anatomical orientation have only been used for the structural evaluation of intramedullary rodding techniques in human femurs (Knothe, 2000). These procedures entailed the removal of an entire diaphyseal segment. Thus, there was no data to compare the femur results that contained such an osteotomy similar to the segment removed in this project.

The results of the mechanical tests were validated before the FEA models were accepted as accurate models. The results show the failure of the whole femur was located at the neck (Appendix N); this is a common location of failure for such loading on femurs (Janice Lalikos, M.D., and Raymond Dunn, M.D, clinical advisors). The failure of the harvested femur bone was located at the proximal cut of the harvested site and not at the distal cut. This location of failure was validated through a simplified analytical stress analysis of the structure. The uniaxial compression and the orientation of the femur cause a bending moment in the shaft of the femur.

The bending moment is higher in the proximal cut as compared to the distal cut. The proximal cut has a larger lever arm (blue) for it further away from the axis the uniaxial force is applied on (red), as can be seen in figure 7.4. This larger lever arm causes a higher moment at the proximal cut; thus, causing an increase stress at this location which causes it to fail before the distal cut.

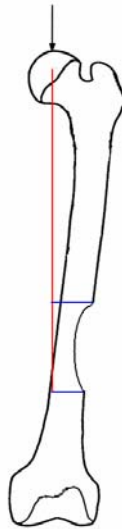


Figure 7.4- Lever Arm - Lever arms created by the orientation of the femur

Sigma Plot 9.0 was used to compare the results from the compression tests. T-test was used to compare the femur structure results in order to test whether the strength of the femur structure is significantly affected when the 35% femur flap or the 40% femur is removed. The first t-test was run to test the hypothesis that the maximum force that the femur can take when loaded in anatomical position under uni-axial compression is significantly affected when the 35% bone segment is removed from the femur. The maximum compressive force of the whole femur is 3980 ± 1330 N and the maximum compressive force of the 35% harvested femur is 3600 ± 1050 N. A 95% confidence interval shows that the whole femurs and the 35% harvested femurs are not significantly different ($P= 0.847$, $n= 4$ whole femurs, $n= 4$ 35% Harvested femurs). Thus, the 35% femur flap removal is not proven to significantly affect the femur structure. The same test was run to test that the 40% harvested femur's structural strength is less than that of the whole femur's structural strength. The maximum compressive force of the 40% harvested femur is 2790 ± 1700 N. The t-test, with the confidence limit set to $P=0.05$, shows that the 40% harvested femurs' structural strengths are not significantly different from the whole femurs' ($P= 0.670$, $n= 4$ whole femurs, $n= 4$ 40% harvested femurs). In conclusion, the removal

of the 40% femur flaps does not significantly affect the femur structure. Refer to Statistical Analysis Results in the Appendix H

Chapter 8: Conclusion

In conclusion, the 40% femur flap from the anteriolateral region of the contralateral femur would be the optimal replacement for the fibula flap, as it can withstand a significantly larger load in 3-point bending. This characteristic would help minimize failures due to mechanical stresses during and after tibial reconstructive surgery. However, the removal of the 40% femoral flap initiates structural yielding in the femur loads lower than the factor of safety used (50th percentile male in stair descent, 2447N). This result leads to the conclusion that a fixation device, such as intramedullary rodding, should be used to support the weakened structure. If fixation of the femur shows to be cost efficient to the patient and time efficient to the surgeon, as compared to the benefits of using a fibular flap (i.e. 225N higher ultimate tensile strength in bending, and a more equivalent elastic modulus to the tibia), this procedure may replace the use of fibular flaps for tibial defect reconstruction and other similar bone defects.

Chapter 9: Recommendations

Due to numerous constraints in this preclinical validation project, there are many suggestions for future continuations in this area of study. The most significant constraint was that the bone samples were from cadavers aging 77-95 years of age and split 50:50 in the gender ratio. Optimal bone samples would be males in their mid-thirties, as this would be better applied to the 50th percentile male in stair descent be more successfully applied to the analysis. Furthermore, the sample size was very small at a total of 16 femurs. To observe greater statistical prevalence, sample size around 30 femurs would be sufficient. Due to time constraints, 3-point bending experiments were not performed on tibial segments. This additional testing would allow for a comparison between the fibular and femoral flaps' ability to match the properties of the tibia. For future studies, higher resolution images should be taken so that bone densities may be used in the calculations. Research on structure of the harvested femur in dynamic and kinematic loads of ambulation were not carried out due to complexity of such research hardware and the time constraints of this project.

Appendices

Appendix A

Weighted Specifications List and Results

Specifications List				
Objectives (0-least important, 6-most important)	Surgical Advisor ~70%	Project Advisor ~30%	Totals Surgical	Totals Project
Taken from vastus intermedialis origin on femur	6	3	4.2	0.9
Stabilize the tibia using a single femoral flap	1	6	0.7	1.8
Tolerate higher loads than the fibular flap	2	5	1.4	1.5
Used for reconstruction of tibial fractures > 6 cm	4	4	2.8	1.2
Match current osteotomy techniques	3	2	2.1	0.6
Must not compromise the donating femur's structure	5	1	3.5	0.3

Combined Totals of UMMS and Prof. Billiar	
Objectives (0-least important, 6-most important)	Totals
Taken from vastus intermedialis origin on femur	5.1
Stabilize the tibia using a single femoral flap	2.5
Used for reconstruction of tibial fractures > 6 cm	2.9
Tolerate higher loads than the fibular flap	4
Match current osteotomy techniques	2.7
Must not compromise the donating femur's structure	3.8

Weighted Objectives List in Order of Importance
Taken from vastus intermedialis origin on femur
Tolerate higher loads than the fibular flap
Must not compromise the donating femur's structure
Used for reconstruction of tibial fractures > 6 cm
Match current osteotomy techniques
Stabilize the tibia using a single femoral flap

Appendix B Femur/Fibula Experimentation Pairwised Comparison Chart

Testing Options	simplehardware (18.2%)	frequentlyused (26.2%)	oncampus (6.2%)	easymachine (2.2%)	easyresults (10.2%)	simulatesanatomy (14.2%)	biomechpref (22.2%)
Shear Test	0	1	1	1	1	1	1
Compression Test	1	0	1	1	1	0	0
Tensile Test	1	0	1	1	1	0	0
3-Point Bending Test	1	1	1	1	1	1	1
4-Point Bending Test	0	1	1	1	1	1	1
Combined Shear-Tensile Test	0	0	1	0	0	0	0
Combined Shear-Compression Test	0	1	1	0	0	1	1

Testing Options	Totals
Shear Test	0.812
Compression Test	0.368
Tensile Test	0.368
3-Point Bending Test	0.994
4-Point Bending Test	0.812
Combined Shear-Tensile Test	0.062
Combined Shear-Compression Test	0.688
Testing Options in Order of Efficiency	
3-Point Bending Test	
4-Point Bending Test	
Shear Test	
Combined Shear-Compression Test	
Compression Test	
Tensile Test	
Combined Shear-Tensile Test	

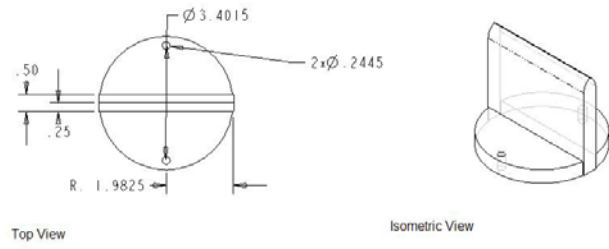
Appendix C

PCC for 3-Point Bending Abutments

	Long Washer	Thin Washer	3-Point Grip	3-Point Skid
2.1.1. Abutment Design Objectives				
The abutment design must be...				
· Capable of holding a cylinder shaped bone segment	1	1	1	1
· Capable of holding a wedge shaped bone segment	1	1	1	1
· Holds a 10cm long bone segment	1	1	1	1
· Structured for easy view of specimen during testing and accessible	1	1	1	1
· User friendly	1	1	1	1
· Easy to manufacture	0	0	0	1
2.1.1. Abutment Design Constraints				
· Must not apply initial load to specimen	1	1	1	1
· Size compatibility with fibula	1	1	1	1
· Size compatibility with femur wedge	1	1	1	1
· Size compatibility with tibia	1	1	1	1
· Inexpensive to manufacture	0	0	0	1
· Minimizes data acquisition complications	0	0	0	1
· Compatible with Worcester Polytechnic Institute materials testing devices	1	1	1	1
2.1.2. Abutment Design Functions				
· Applies loads at 25%, 50%, and 75% of the length of the bone	1	1	1	1
· Must withstand loads up to 50 lbs (222 Newtons)	1	1	1	1
· Distributes loads into the bone structure with minimal stress focus points	0	1	0	1
· Fixes bone segments in one position, not allowing it to move as forces are applied	1	1	1	1
TOTALS:	13	14	13	17

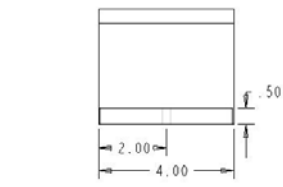
Appendix D

3-Point Bending Hardware

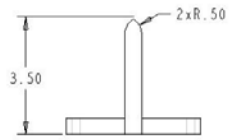


Top View

Isometric View



Front View



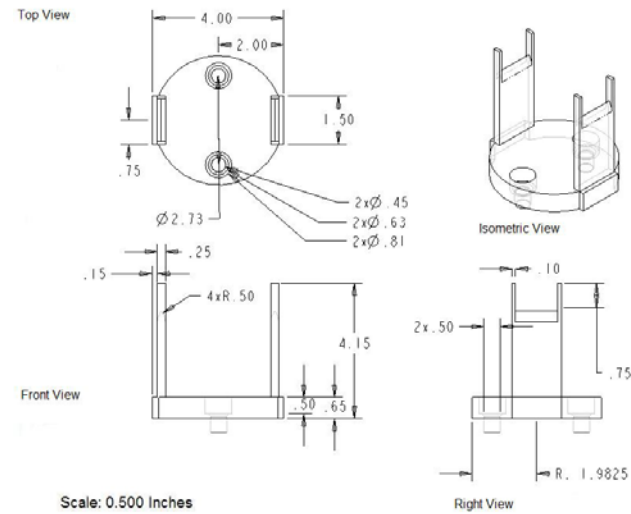
Right View

Scale: 0.500 Inches

Part: Top 3-Point Bending Abutment

Date: 12/7/04

By: BME Femur Flap MQP '04



Top View

Isometric View

Front View

Right View

Scale: 0.500 Inches

Part: Bottom 3-Point Bending Abutment

Date: 12/7/04

By: BME Femur Flap MQP '04

Appendix E

Femur Host Experimentation Pairwised Comparison Chart

Testing Options	simplehardware (21.9%)	frequentlyused (29.9%)	oncampus (9.9%)	easymachine (5.9%)	easyresults (13.9%)	simulatesanatomy (17.9%)
Dynamic Loading	0	1	0	1	1	1
Compression Test	1	1	1	1	1	0
Anatomical Compression	1	1	1	1	1	1
3-Point Bending Test	1	1	1	1	1	0
Tension Test	0	0	1	1	1	0

Testing Options	Totals
Dynamic Loading	0.676
Compression Test	0.815
Anatomical Compression	0.994
3-Point Bending Test	0.815
Tension Test	0.297
Testing Options in Order of Efficiency	
Anatomical Compression	
3-Point Bending Test	
Compression Test	
Dynamic Loading	
Tension Test	

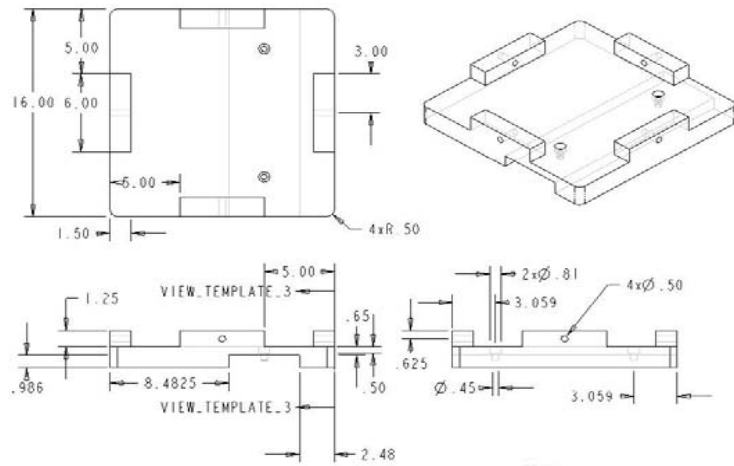
Appendix F

PCC for Compression Abutments

	Blocks	Triangles	Tree Stand A	Design B	Free Tree Stand	Fixed Free Tree Stand
2.1.1. Abutment Design Objectives						
The abutment design must be...						
• Capable of accommodating anatomical orientation of the femur	1	1	1	1	1	1
• Adjustable for different bone sizes	0	0	1	1	1	1
• Capable of fastening bone samples	1	1	1	1	0	1
• Structured for easy view of specimen during testing and accessible	1	1	1	1	1	1
• User friendly	0	0	0	0	1	1
• Easy to manufacture	1	1	0	0	1	1
2.1.1. Abutment Design Constraints						
• Must not apply initial load to specimen	1	1	1	1	1	1
• Size compatibility with femoral specimen and materials testing devices	1	1	1	1	1	1
• Inexpensive to manufacture	1	1	0	0	1	1
• Minimizes data acquisition complications	0	0	0	0	0	1
• Compatible with Worcester Polytechnic Institute materials testing devices	1	1	0	0	1	1
2.1.2. Abutment Design Functions						
• Applies loads to femur with an orientation of an 8°-11° anterior angle and a 7° angle of adduction.	0	0	1	1	0	1
• Must withstand loads up to 550 lbs (2430 Newtons)	1	0	1	1	1	1
• Adjusts to different lengths of femurs in order to maintain physiological orientation.	0	0	1	1	1	1
• Distributes loads into the bone structure with minimal stress focus points.	1	1	1	1	1	1
• Fixes the femur in one position, not allowing it to move as forces are applied.	1	1	1	1	0	1
TOTALS:	11	10	11	11	12	16

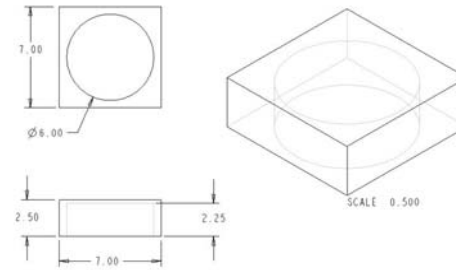
Appendix G

Compression Hardware

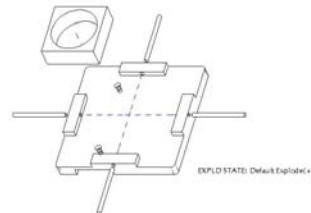
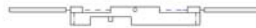


Scale: 0.200 Inches
 Part: Compressive Abutment Base
 Date: 12/6/04
 By: BME Femur Flap MQP '04

Note:
 Counter-Bored screw holes
 1/2-20 UNF-2B Tab:1.09
 29/64 DnH (0.45) Thru (1) Hole



Scale: 0.300 Inches
 Part: Cup Holder
 Date: 12/7/04
 By: BME Femur Flap MQP '04



Appendix H

Statistical Procedure

1. First, check for outliers in the data
 - a. Go to the *start* menu, select *Programs* and go to *SPSS 12.0 for windows*
 - b. To start a new file, type the data directly onto the worksheet, or import it from Excel by selecting *Import data* from the *File* menu and select the data you would like to import. The data must be set up in the following way:

Factor 1 (for blocking data)	Factor 2 (for blocking data)	Data
<i>Enter data for factor 1</i>	<i>Enter data for factor 2</i>	<i>Enter numerical data</i>

- c. Go to the “Graph” pull-down menu and select “*Q-Q*” to plot the data and check for outliers using the residual and nominal values
 - d. In the “Q-Q Plots” window that pops up select the following items:
 - i. Select “*Variables*”, to name the variables for comparison
 - ii. Under “*Transform*” select *standardize values*
 - iii. Set “*Test Distribution*” to *normal*
 - iv. Select the “*Proportion Estimation Formula*” that you would like to use in this test (*Tukey’s* was selected for this particular experiment)
 - v. Select the “*Rank Assigned to Means*” (*Means* was chosen as the rank in this experiment)
 - vi. Click finish and the analysis will be run to check for outliers
 - vii. Remove all outliers from the data
2. Then, run statistical analysis on the data, with the outliers removed
 - a. Go to the *start* menu, select *Programs*, go to *SigmaPlot* and select *SigmaPlot 9.0*
 - b. To start a new file, type the data directly onto the worksheet, or import it from Excel by selecting *Import data* from the *File* menu and select the data you would like to import. The data must be set up in the following way:

Factor 1 (for blocking data)	Factor 2 (for blocking data)	Data
<i>Enter data for factor 1</i>	<i>Enter data for factor 2</i>	<i>Enter numerical data</i>

- c. To compare data with two factors:
 - i. Go to the *Statistics* tab and select “*Two Way ANOVA*”. Two way ANOVA is used to block the data so that it can be grouped according to factors, which may affect the results, and then analyzed
 - ii. Fill in the “ANOVA” window that pops up:
 1. “Factor A”: select the first column in your data (your “Factor 1”)
 - “Factor B”: select the second column in your data (your “Factor 2”)
 - “Data”: select the third column in your data (your numerical “Data”)
 - Click “OK”
 2. The “Multiple Comparison Options” window will pop up

- a. Select the type of test under the “Suggested Test” pull-down menu
 - i. Select *Tukey* for all pairwise comparisons of the mean responses to different treatment groups
 - ii. Select *Fisher LSD* if you want additional analysis done
 - b. Under “Factors to compare”, select the factors that you want to compare
 - c. For “Comparison Type” choose *all pairwise*, since the data is blocked
 - d. Click “OK” and the statistical analysis is run.
- d. To compare two groups:
- i. Go to the *Statistics* tab, select “*Compare Two Groups*” and then select “*t-test*”. The t-test is used to compare 2 groups to see if one is greater than another.
 - ii. Fill in the “Pick Columns for t-test” window that pops up:
 1. Select the “*Data format*” tab and choose “*Indexed*” to place the groups or treatments from the data in a factor column and the data points in a second column.
 2. Select the “Factor” column in your data that would want to do analysis on and plug it in for “*Data*”
 3. Select the raw data column and plug it in for the second “*Data*” set
 4. Click “*Finish*” and the statistical analysis is run
 - a. If the normality test or the equal variance test fails, run *the Mann-Whitney Rank Sum Test*

Statistical Analysis Results

Testing for Outliers using Q-Q Plots in SPSS 12.0 for Windows:

35% Femur Flaps Tested for Outliers:

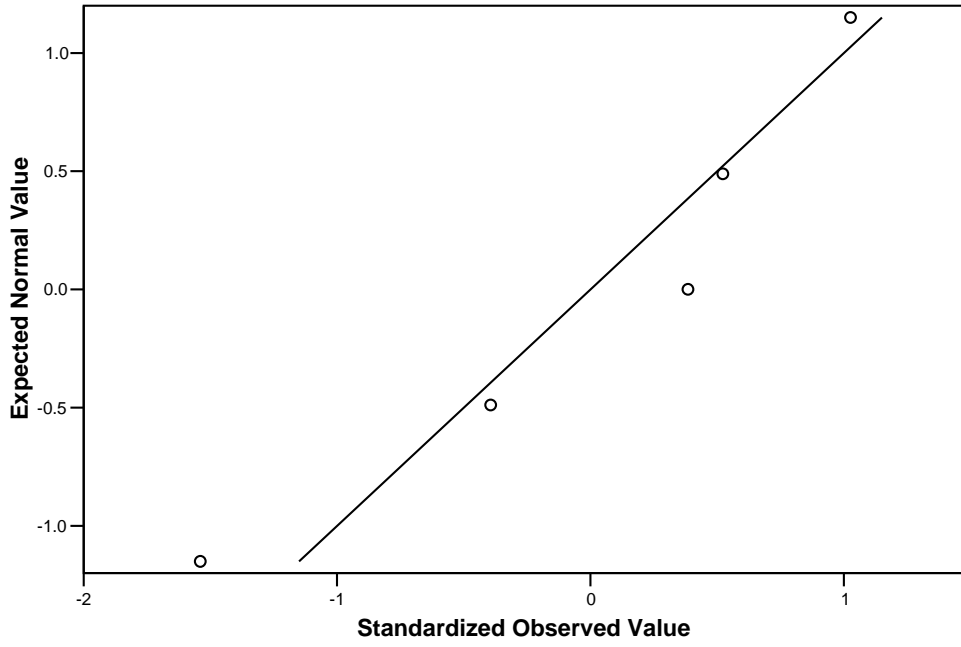
PPlot

```
MODEL:  MOD_1.
Distribution tested: Normal
Proportion estimation formula used: Tukey's
Rank assigned to ties: Mean
```

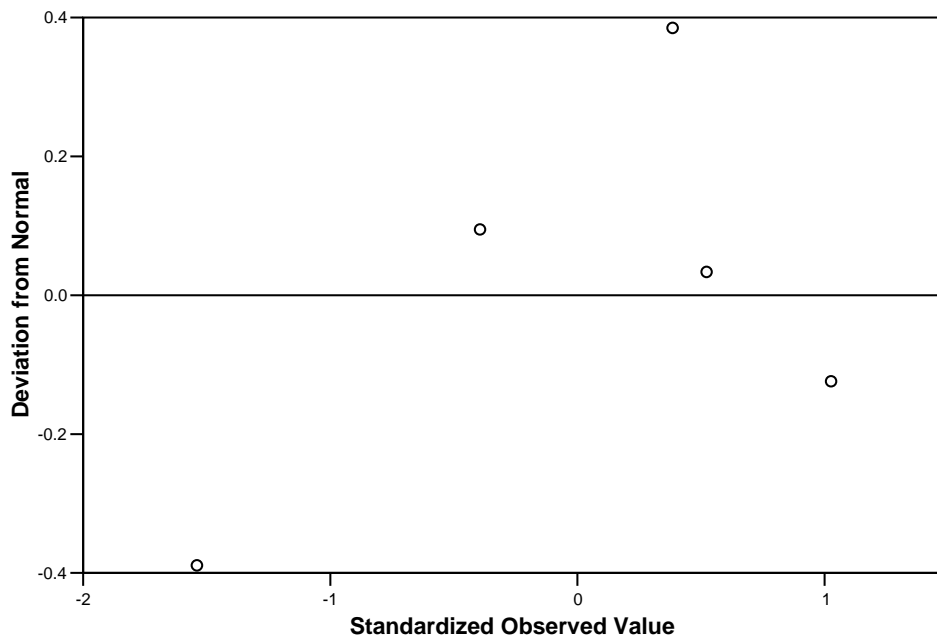
—

```
For variable Specimen ...
Normal distribution parameters estimated: location = 0 and scale = 1
For variable ForceN ...
Normal distribution parameters estimated: location = 0 and scale = 1
```

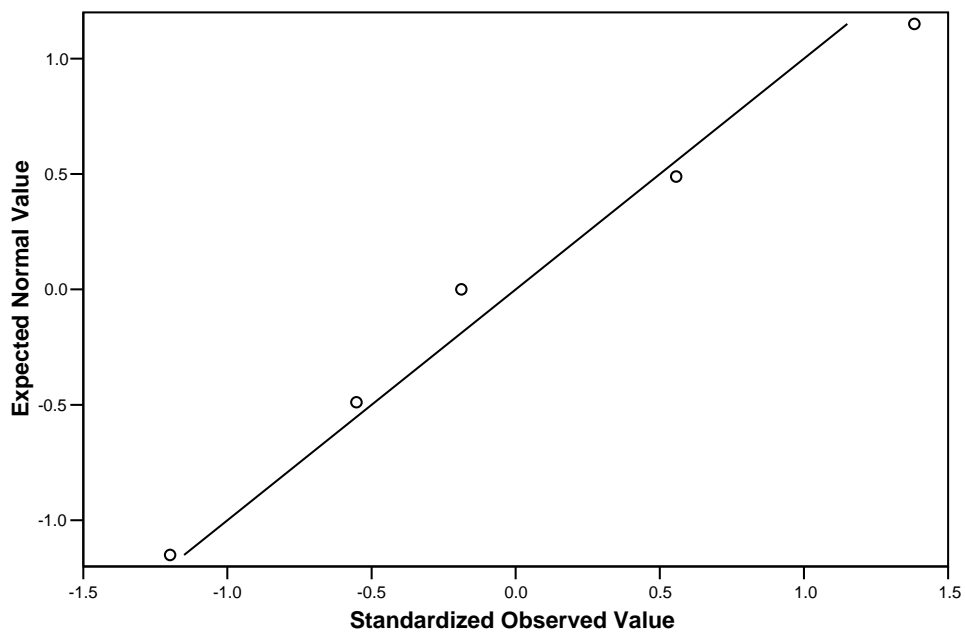
Normal Q-Q Plot of Specimen



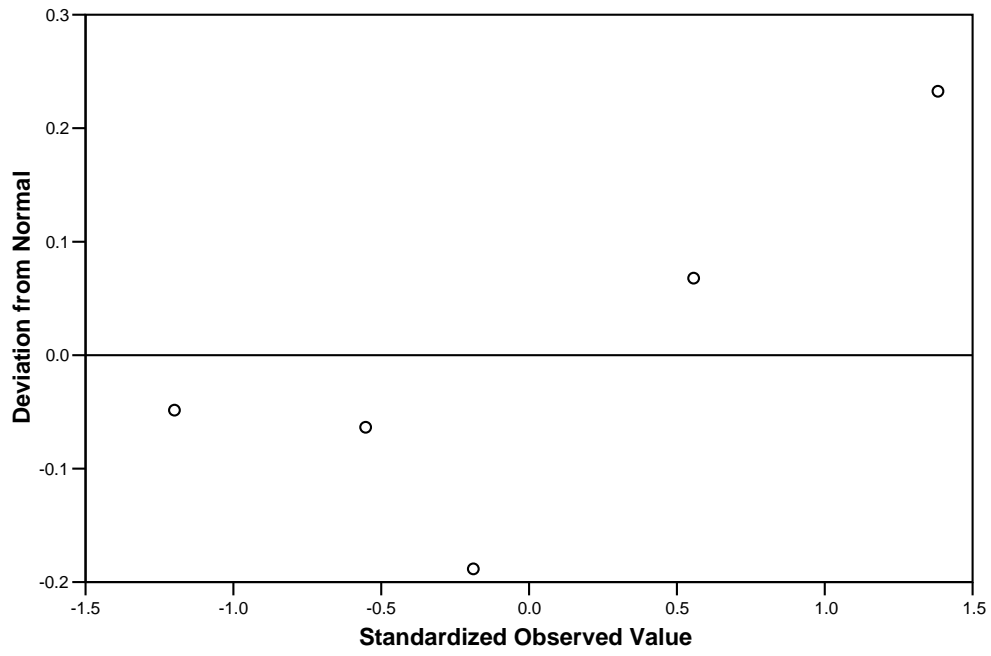
Detrended Normal Q-Q Plot of Specimen



Normal Q-Q Plot of Force (N)



Detrended Normal Q-Q Plot of Force (N)



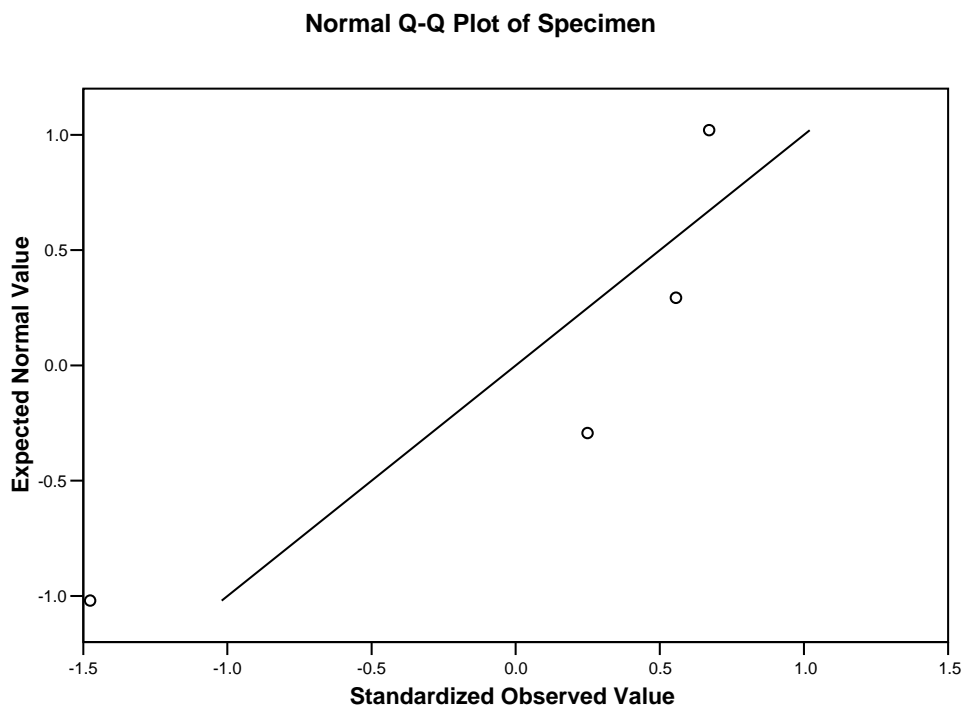
40% Femur Flaps Tested for Outliers:

PPlot

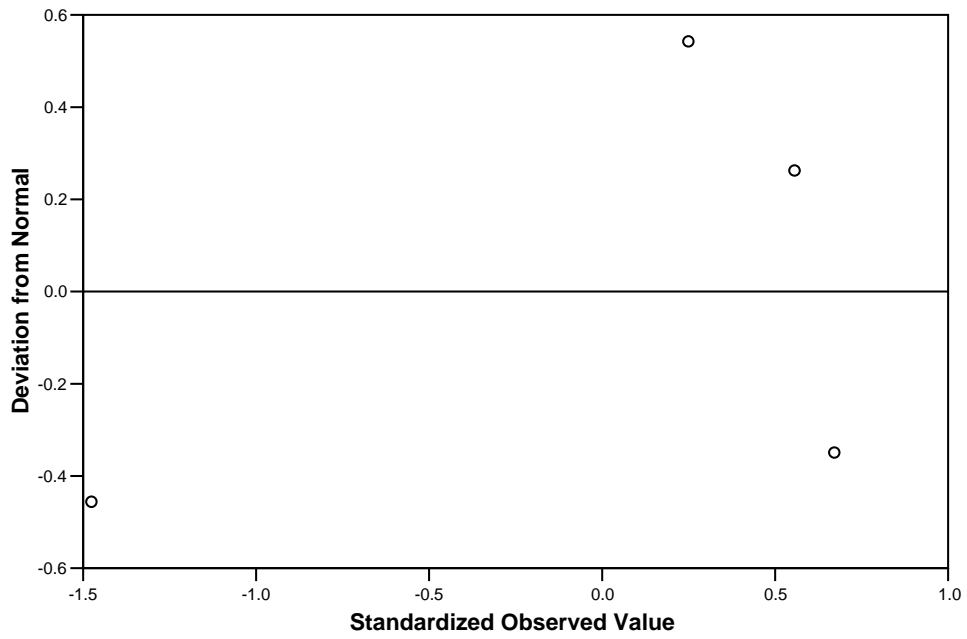
MODEL: MOD_3.
Distribution tested: Normal
Proportion estimation formula used: Tukey's
Rank assigned to ties: Mean

—

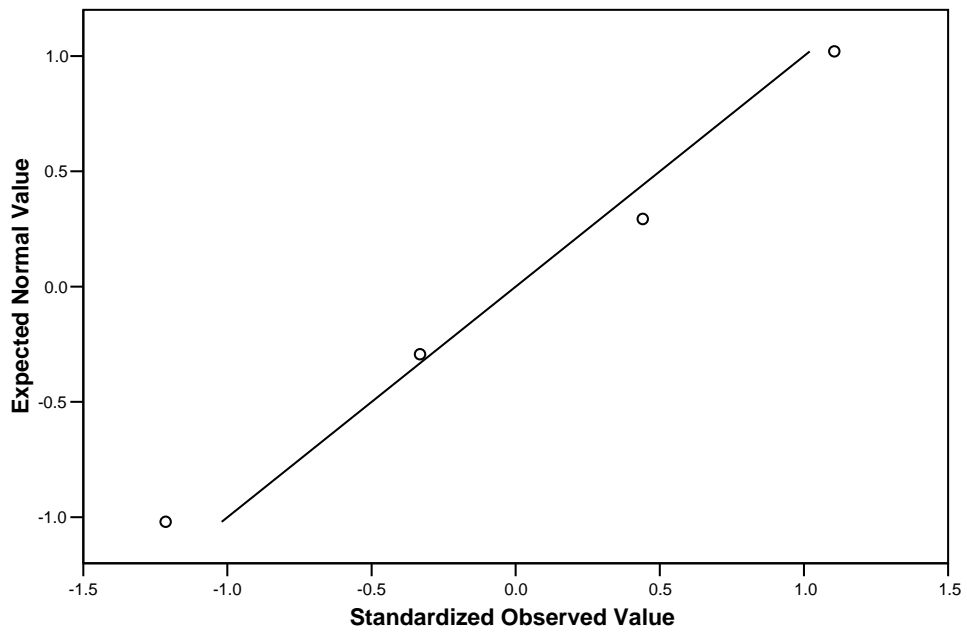
For variable Specimen ...
Normal distribution parameters estimated: location = 0 and scale = 1
For variable ForceN ...
Normal distribution parameters estimated: location = 0 and scale = 1



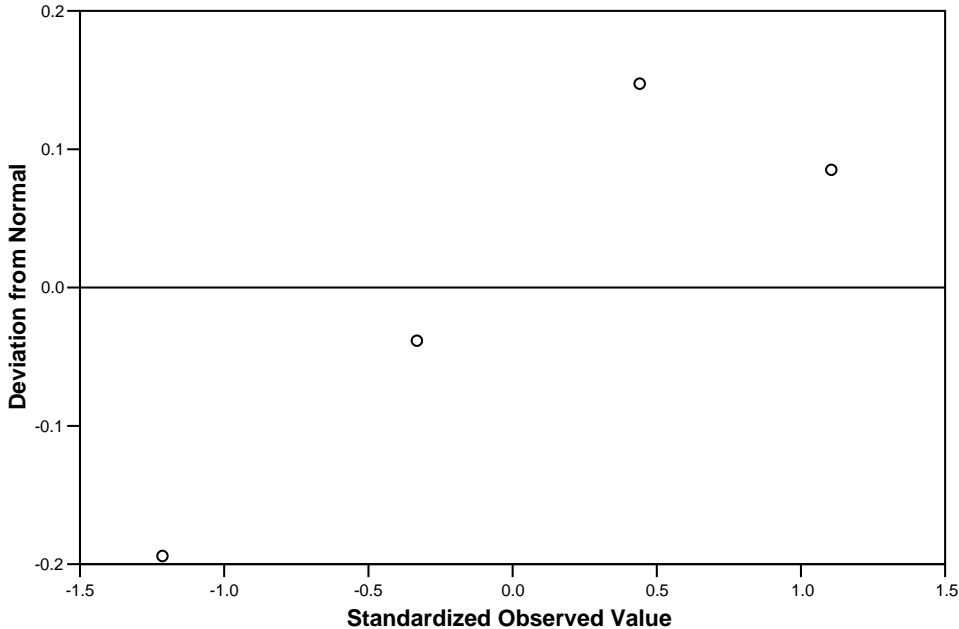
Detrended Normal Q-Q Plot of Specimen



Normal Q-Q Plot of Force (N)



Detrended Normal Q-Q Plot of Force (N)



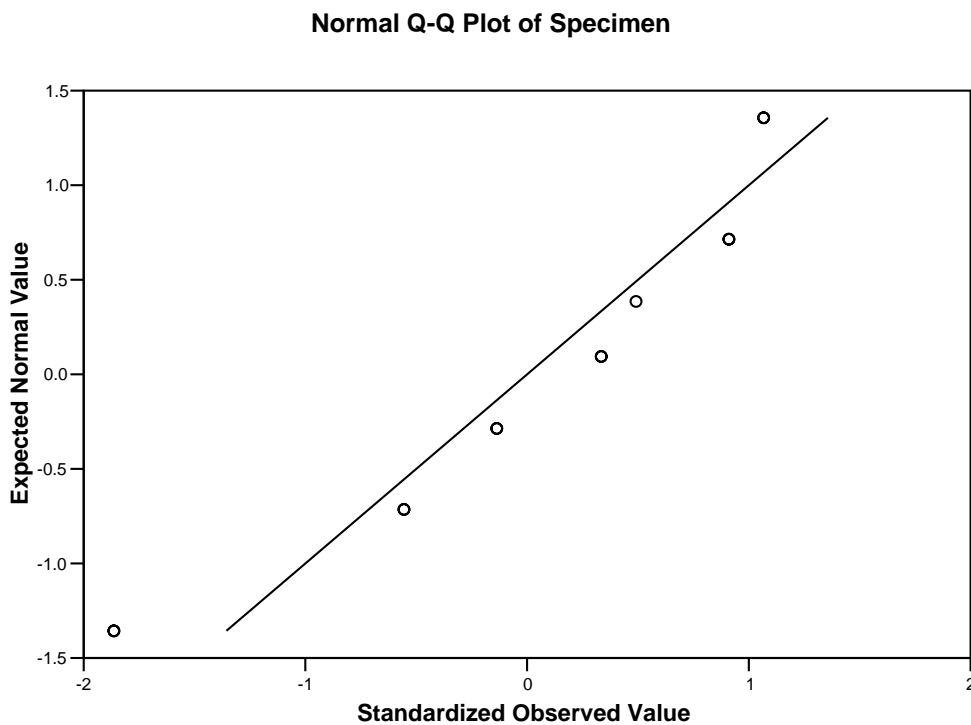
Fibula Flaps Tested for Outliers:

PPlot

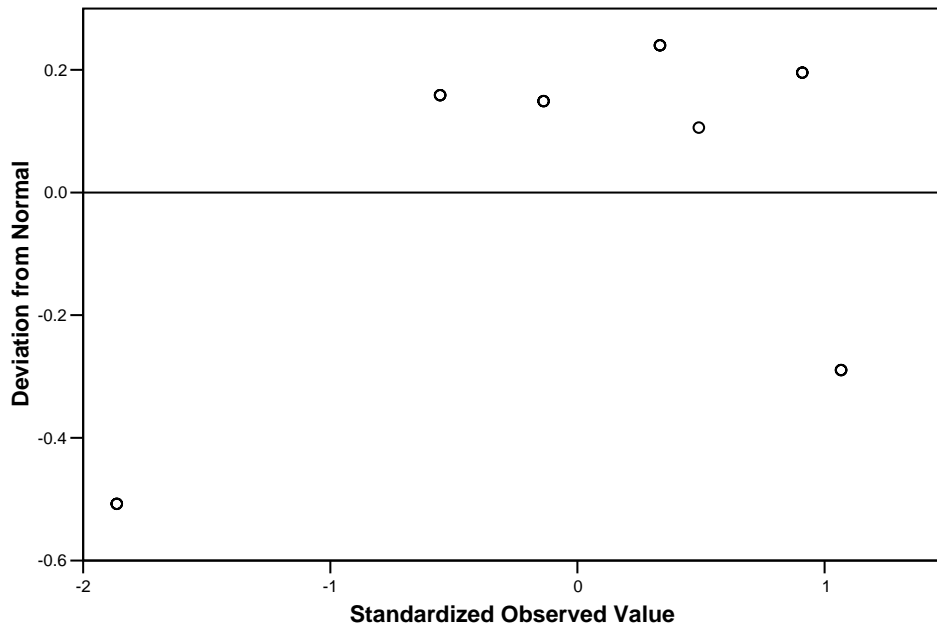
MODEL: MOD_4.
Distribution tested: Normal
Proportion estimation formula used: Tukey's
Rank assigned to ties: Mean

—

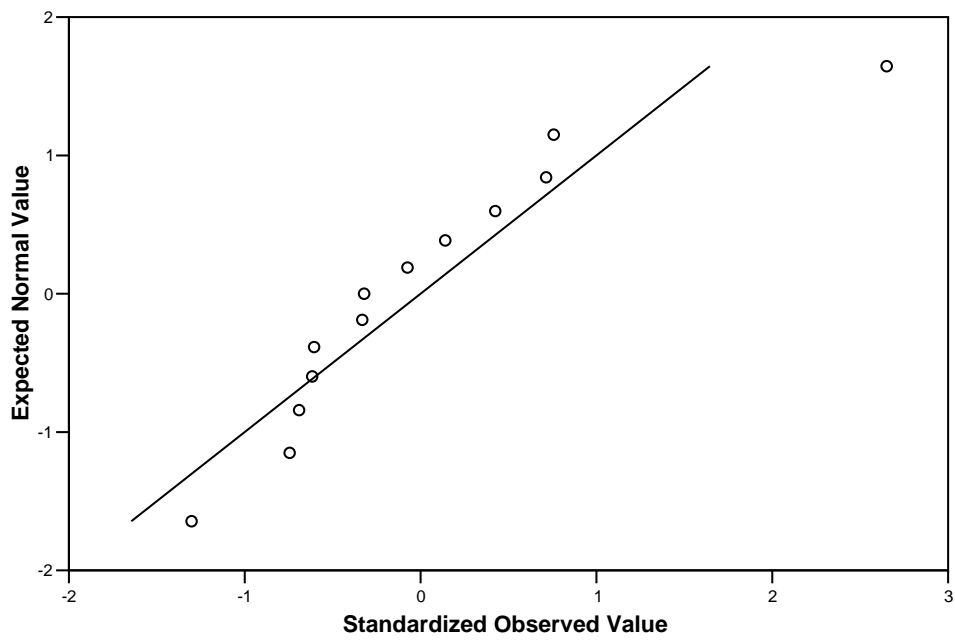
For variable Specimen ...
Normal distribution parameters estimated: location = 0 and scale = 1
For variable ForceN ...
Normal distribution parameters estimated: location = 0 and scale = 1



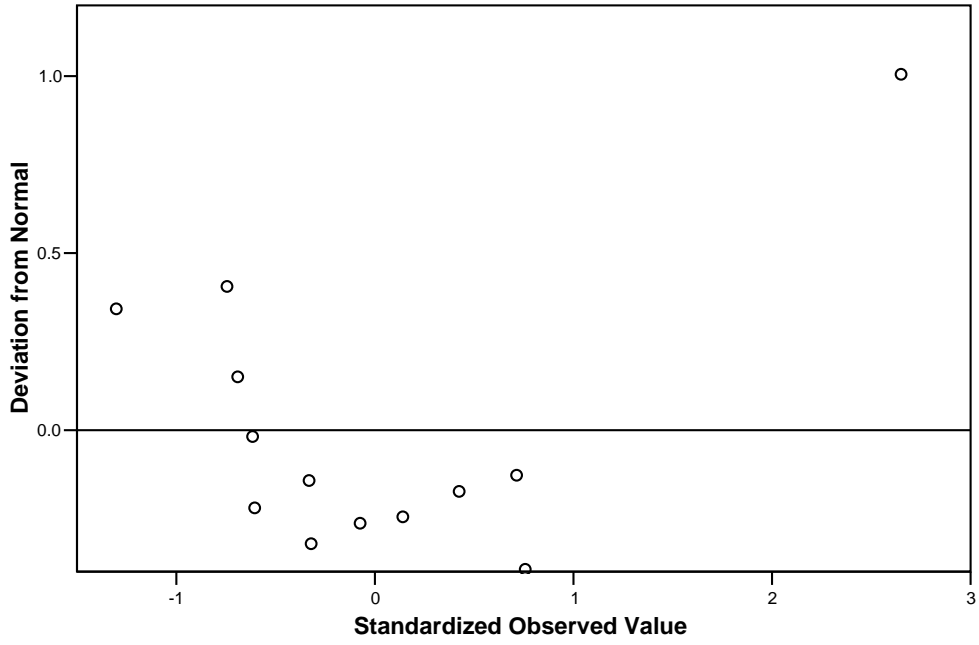
Detrended Normal Q-Q Plot of Specimen



Normal Q-Q Plot of Force (N)



Detrended Normal Q-Q Plot of Force (N)



Testing for Significant Results:

Table of 3-point bending results used for Sigma Plot 9.0 in comparing the 35% Femur Flaps and the Fibula Flaps:

Specimen	Flap of Femur 35 or Fibula	Force (N)
1975	Femur	590
1975	Fibula	511
1975	Fibula	383
2000	Femur	948
2000	Fibula	612
2000	Fibula	705
2017	Femur	771
2017	Fibula	564
2017	Fibula	487
2020	Femur	1240
2020	Fibula	1120
2031	Femur	797
2031	Fibula	566
2031	Fibula	767
2008	Fibula	759
2008	Fibula	652

Comparison of 35% femur flaps versus fibula flaps using the Tukey Test:

Two Way Analysis of Variance

Data source: Data 1 in 35%Flaps_Tukey

General Linear Model (No Interactions)

Dependent Variable: Force (N)

Normality Test: Passed (P = 0.379)

Equal Variance Test: Failed (P < 0.050)

Source of Variation	DF	SS	MS	F	P
Specimen	5	562692.182	112538.436	17.977	<0.001
Flap of Femur 35 or Fibula	1	113305.579	113305.579	18.100	0.002
Residual	9	56340.254	6260.028		
Total	15	787504.000	52500.267		

The difference in the mean values among the different levels of Specimen is greater than would be expected by chance after allowing for effects of differences in Flap of Femur 35 or Fibula. There is a statistically significant difference (P = <0.001). To isolate which group(s) differ from the others use a multiple comparison procedure.

The difference in the mean values among the different levels of Flap of Femur 35 or Fibula is greater than would be expected by chance after allowing for effects of differences in Specimen. There is a statistically significant difference ($P = 0.002$). To isolate which group(s) differ from the others use a multiple comparison procedure.

Power of performed test with $\alpha = 0.0500$: for Specimen : 1.000

Power of performed test with $\alpha = 0.0500$: for Flap of Femur 35 or Fibula : 0.965

Least square means for Specimen :

Group	Mean	SEM
1975.000	526.193	46.277
2000.000	786.526	46.277
2017.000	638.860	46.277
2020.000	1180.000	55.947
2031.000	741.526	46.277
2008.000	800.079	60.202

Least square means for Flap of Femur 35 or Fibula :

Group	Mean	SEM
2.000	873.443	36.866
1.000	684.285	24.296

All Pairwise Multiple Comparison Procedures (Tukey Test):

Comparisons for factor: **Specimen**

Comparison	Diff of Means	p	q	P	P<0.050
2020.000 vs. 1975.000	653.807	6	12.735	<0.001	Yes
2020.000 vs. 2017.000	541.140	6	10.540	<0.001	Yes
2020.000 vs. 2031.000	438.474	6	8.541	0.002	Yes
2020.000 vs. 2000.000	393.474	6	7.664	0.004	Yes
2020.000 vs. 2008.000	379.921	6	6.538	0.011	Yes
2008.000 vs. 1975.000	273.886	6	5.101	0.046	Yes
2008.000 vs. 2017.000	161.219	6	3.003	0.355	No
2008.000 vs. 2031.000	58.553	6	1.091	0.966	Do Not Test
2008.000 vs. 2000.000	13.553	6	0.252	1.000	Do Not Test
2000.000 vs. 1975.000	260.333	6	5.626	0.027	Yes
2000.000 vs. 2017.000	147.667	6	3.191	0.301	Do Not Test
2000.000 vs. 2031.000	45.000	6	0.972	0.979	Do Not Test
2031.000 vs. 1975.000	215.333	6	4.653	0.073	No
2031.000 vs. 2017.000	102.667	6	2.219	0.635	Do Not Test
2017.000 vs. 1975.000	112.667	6	2.435	0.551	Do Not Test

Comparisons for factor: **Flap of Femur 35 or Fibula**

Comparison	Diff of Means	p	q	P	P<0.050
2.000 vs. 1.000	189.158	2	6.059	0.002	Yes

A result of "Do Not Test" occurs for a comparison when no significant difference is found between two means that enclose that comparison. For example, if you had four means sorted in order, and found no difference between means 4 vs. 2, then you would not test 4 vs. 3 and 3 vs. 2, but still test 4 vs. 1 and 3 vs. 1 (4 vs. 3 and 3 vs. 2 are enclosed by 4 vs. 2: 4 3 2 1). Note that not testing the enclosed means is a procedural rule, and a result of Do Not Test should be treated as if there is no significant difference between the means, even though one may appear to exist.

Table of 3-point bending results used for Sigma Plot 9.0 in comparing the 35% Femur Flaps, with a safety factor or 100N, and the Fibula Flaps:

Specimen	Flap of Femur 35 or Fibula	Force (N)	Femur Force Minus 100 N (N)
1975	Femur	490	590-100
1975	Fibula	511	
1975	Fibula	383	
2000	Femur	848	948-100
2000	Fibula	612	
2000	Fibula	705	
2017	Femur	671	771-100
2017	Fibula	564	
2017	Fibula	487	
2020	Femur	1140	1240-100
2020	Fibula	1120	
2031	Femur	697	797-100
2031	Fibula	566	
2031	Fibula	767	
2008	Fibula	759	
2008	Fibula	652	

Comparison of 35% femur flaps, taking into account the 100N safety factor, versus fibula flaps using the Least Square Difference:

Two Way Analysis of Variance

Data source: Data 1 in Flaps_MINUS100_LSD

General Linear Model (No Interactions)

Dependent Variable: Force (N)

Normality Test: Passed (P = 0.379)

Equal Variance Test: Failed (P < 0.050)

Source of Variation	DF	SS	MS	F	P
Specimen	5	562692.182	112538.436	17.977	<0.001
Flap of Femur 35 or Fibula	1	25172.246	25172.246	4.021	0.076
Residual	9	56340.254	6260.028		
Total	15	669679.000	44645.267		

The difference in the mean values among the different levels of Specimen is greater than would be expected by chance after allowing for effects of differences in Flap of Femur 35 or Fibula. There is a statistically significant difference (P = <0.001). To isolate which group(s) differ from the others use a multiple comparison procedure.

The difference in the mean values among the different levels of Flap of Femur 35 or Fibula is not great enough to exclude the possibility that the difference is just due to random sampling variability after allowing for the effects of differences in Specimen. There is not a statistically significant difference (P = 0.076).

Power of performed test with alpha = 0.0500: for Specimen : 1.000
 Power of performed test with alpha = 0.0500: for Flap of Femur 35 or Fibula : 0.332

Least square means for Specimen :

Group	Mean	SEM
1975.000	476.193	46.277
2000.000	736.526	46.277
2017.000	588.860	46.277
2020.000	1130.000	55.947
2031.000	691.526	46.277
2008.000	750.079	60.202

Least square means for Flap of Femur 35 or Fibula :

Group	Mean	SEM
2.000	773.443	36.866
1.000	684.285	24.296

All Pairwise Multiple Comparison Procedures (Fisher LSD Method):

Comparisons for factor: **Specimen**

Comparison	Diff of Means	LSD(alpha=0.050)	P	Diff >= LSD
2020.000 vs. 1975.000	653.807	164.246	<0.001	Yes
2020.000 vs. 2017.000	541.140	164.246	<0.001	Yes
2020.000 vs. 2031.000	438.474	164.246	<0.001	Yes
2020.000 vs. 2000.000	393.474	164.246	<0.001	Yes
2020.000 vs. 2008.000	379.921	185.914	0.001	Yes
2008.000 vs. 1975.000	273.886	171.772	0.006	Yes
2008.000 vs. 2017.000	161.219	171.772	0.063	No
2008.000 vs. 2031.000	58.553	171.772	0.460	Do Not Test
2008.000 vs. 2000.000	13.553	171.772	0.862	Do Not Test
2000.000 vs. 1975.000	260.333	148.049	0.003	Yes
2000.000 vs. 2017.000	147.667	148.049	0.050	Do Not Test
2000.000 vs. 2031.000	45.000	148.049	0.509	Do Not Test
2031.000 vs. 1975.000	215.333	148.049	0.009	Yes
2031.000 vs. 2017.000	102.667	148.049	0.151	Do Not Test
2017.000 vs. 1975.000	112.667	148.049	0.119	No

Comparisons for factor: **Flap of Femur 35 or Fibula**

Comparison	Diff of Means	LSD(alpha=0.050)	P	Diff >= LSD
2.000 vs. 1.000	89.158	99.879	0.074	No

A result of "Do Not Test" occurs for a comparison when no significant difference is found between two means that enclose that comparison. For example, if you had four means sorted in order, and found no difference between means 4 vs. 2, then you would not test 4 vs. 3 and 3 vs. 2, but still test 4 vs. 1 and 3 vs. 1 (4 vs. 3 and 3 vs. 2 are enclosed by 4 vs. 2: 4 3 2 1). Note that not testing the enclosed means is a procedural rule, and a result of Do Not Test should be treated as if there is no significant difference between the means, even though one may appear to exist.

Table of 3-point bending results used for Sigma Plot 9.0 in comparing the 40% Femur Flaps and the Fibula Flaps:

Specimen	Flap of Femur 40 or Fibula	Force (N)
1975	Femur	1010
1975	Fibula	511
1975	Fibula	383
2000	Fibula	612
2000	Fibula	705
2017	Fibula	564
2017	Fibula	487
2020	Femur	1940
2020	Fibula	1120
2031	Femur	1510
2031	Fibula	566
2031	Fibula	767
2008	Fibula	759
2008	Fibula	652
2028	Fibula	497
2028	Fibula	513
2028	Femur	439

Comparison of 40% femur flaps versus fibula flaps using the Tukey Test:

Two Way Analysis of Variance

Data source: Data 1 in 40% flaps vs fibula

General Linear Model (No Interactions)

Dependent Variable: Force (N)

Normality Test: Passed (P = 0.165)

Equal Variance Test: Failed (P < 0.050)

Source of Variation	DF	SS	MS	F	P
Specimen	6	1287178.320	214529.720	4.996	0.016
Flap of Femur 40 or Fibula	1	679818.711	679818.711	15.832	0.003
Residual	9	386460.122	42940.014		
Total	16	2770795.059	173174.691		

The difference in the mean values among the different levels of Specimen is greater than would be expected by chance after allowing for effects of differences in Flap of Femur 40 or Fibula. There is a statistically significant difference (P = 0.016). To isolate which group(s) differ from the others use a multiple comparison procedure.

The difference in the mean values among the different levels of Flap of Femur 40 or Fibula is greater than would be expected by chance after allowing for effects of differences in Specimen. There is a statistically significant difference (P = 0.003). To isolate which group(s) differ from the others use a multiple comparison procedure.

Power of performed test with alpha = 0.0500: for Specimen : 0.767
 Power of performed test with alpha = 0.0500: for Flap of Femur 40 or Fibula : 0.938

Least square means for Specimen :

Group	Mean	SEM
1975.000	721.578	121.616
2000.000	919.233	160.512
2017.000	786.233	160.512
2020.000	1530.000	146.526
2031.000	1034.578	121.616
2008.000	966.233	160.512
2028.000	569.911	121.616

Least square means for Flap of Femur 40 or Fibula :

Group	Mean	SEM
2.000	1193.271	115.033
1.000	671.805	58.461

All Pairwise Multiple Comparison Procedures (Tukey Test):

Comparisons for factor: **Specimen**

Comparison	Diff of Means	p	q	P	P<0.050
2020.000 vs. 2028.000	960.089	7	7.130	0.008	Yes
2020.000 vs. 1975.000	808.422	7	6.004	0.024	Yes
2020.000 vs. 2017.000	743.767	7	4.840	0.075	No
2020.000 vs. 2000.000	610.767	7	3.974	0.174	Do Not Test
2020.000 vs. 2008.000	563.767	7	3.668	0.232	Do Not Test
2020.000 vs. 2031.000	495.422	7	3.679	0.230	Do Not Test
2031.000 vs. 2028.000	464.667	7	3.821	0.201	No
2031.000 vs. 1975.000	313.000	7	2.574	0.567	Do Not Test
2031.000 vs. 2017.000	248.344	7	1.744	0.865	Do Not Test
2031.000 vs. 2000.000	115.344	7	0.810	0.996	Do Not Test
2031.000 vs. 2008.000	68.344	7	0.480	1.000	Do Not Test
2008.000 vs. 2028.000	396.322	7	2.783	0.489	Do Not Test
2008.000 vs. 1975.000	244.656	7	1.718	0.872	Do Not Test
2008.000 vs. 2017.000	180.000	7	1.121	0.980	Do Not Test
2008.000 vs. 2000.000	47.000	7	0.293	1.000	Do Not Test
2000.000 vs. 2028.000	349.322	7	2.453	0.613	Do Not Test
2000.000 vs. 1975.000	197.656	7	1.388	0.946	Do Not Test
2000.000 vs. 2017.000	133.000	7	0.829	0.996	Do Not Test
2017.000 vs. 2028.000	216.322	7	1.519	0.921	Do Not Test
2017.000 vs. 1975.000	64.656	7	0.454	1.000	Do Not Test
1975.000 vs. 2028.000	151.667	7	1.247	0.967	Do Not Test

Comparisons for factor: **Flap of Femur 40 or Fibula**

Comparison	Diff of Means	p	q	P	P<0.050
2.000 vs. 1.000	521.467	2	5.715	0.003	Yes

A result of "Do Not Test" occurs for a comparison when no significant difference is found between two means that enclose that comparison. For example, if you had four means sorted in order, and found no difference between

means 4 vs. 2, then you would not test 4 vs. 3 and 3 vs. 2, but still test 4 vs. 1 and 3 vs. 1 (4 vs. 3 and 3 vs. 2 are enclosed by 4 vs. 2: 4 3 2 1). Note that not testing the enclosed means is a procedural rule, and a result of Do Not Test should be treated as if there is no significant difference between the means, even though one may appear to exist.

Comparison of 40% femur flaps versus fibula flaps using the Least Square Difference:

Two Way Analysis of Variance

Data source: Data 1 in 40% flaps vs fibula

General Linear Model (No Interactions)

Dependent Variable: Force (N)

Normality Test: Passed (P = 0.165)

Equal Variance Test: Failed (P < 0.050)

Source of Variation	DF	SS	MS	F	P
Specimen	6	1287178.320	214529.720	4.996	0.016
Flap of Femur 40 or Fibula	1	679818.711	679818.711	15.832	0.003
Residual	9	386460.122	42940.014		
Total	16	2770795.059	173174.691		

The difference in the mean values among the different levels of Specimen is greater than would be expected by chance after allowing for effects of differences in Flap of Femur 40 or Fibula. There is a statistically significant difference (P = 0.016). To isolate which group(s) differ from the others use a multiple comparison procedure.

The difference in the mean values among the different levels of Flap of Femur 40 or Fibula is greater than would be expected by chance after allowing for effects of differences in Specimen. There is a statistically significant difference (P = 0.003). To isolate which group(s) differ from the others use a multiple comparison procedure.

Power of performed test with alpha = 0.0500: for Specimen : 0.767

Power of performed test with alpha = 0.0500: for Flap of Femur 40 or Fibula : 0.938

Least square means for Specimen :

Group	Mean	SEM
1975.000	721.578	121.616
2000.000	919.233	160.512
2017.000	786.233	160.512
2020.000	1530.000	146.526
2031.000	1034.578	121.616
2008.000	966.233	160.512
2028.000	569.911	121.616

Least square means for Flap of Femur 40 or Fibula :

Group	Mean	SEM
2.000	1193.271	115.033
1.000	671.805	58.461

All Pairwise Multiple Comparison Procedures (Fisher LSD Method):

Comparisons for factor: **Specimen**

Comparison	Diff of Means	LSD(alpha=0.050)	P	Diff >= LSD
2020.000 vs. 2028.000	960.089	430.764	<0.001	Yes

Table of 3-point bending results used for Sigma Plot 9.0 in comparing the 40% Femur Flaps, with a safety factor or 100N, and the Fibula Flaps:

Specimen	Flap of Femur 40 or Fibula	Force (N)	Femur Force Minus 100 N (N)
1975	Femur	910	1010-100
1975	Fibula	511	
1975	Fibula	383	
2000	Fibula	612	
2000	Fibula	705	
2017	Fibula	564	
2017	Fibula	487	
2020	Femur	1840	1940-100
2020	Fibula	1120	
2031	Femur	1410	1510-100
2031	Fibula	566	
2031	Fibula	767	
2008	Fibula	759	
2008	Fibula	652	
2028	Fibula	497	
2028	Fibula	513	
2028	Femur	339	439-100

Comparison of 40% femur flaps, with 100N safety factor, versus fibula flaps using the Tukey Test:

Two Way Analysis of Variance

Data source: Data 1 in 40% flaps vs fibula

General Linear Model (No Interactions)

Dependent Variable: Force Minus 100 N (Femur Alone)

Normality Test: Passed (P = 0.165)

Equal Variance Test: Failed (P < 0.050)

Source of Variation	DF	SS	MS	F	P
Specimen	6	1287178.320	214529.720	4.996	0.016
Flap of Femur 40 or Fibula	1	444085.378	444085.378	10.342	0.011
Residual	9	386460.122	42940.014		
Total	16	2434995.059	152187.191		

The difference in the mean values among the different levels of Specimen is greater than would be expected by chance after allowing for effects of differences in Flap of Femur 40 or Fibula. There is a statistically significant difference (P = 0.016). To isolate which group(s) differ from the others use a multiple comparison procedure.

The difference in the mean values among the different levels of Flap of Femur 40 or Fibula is greater than would be expected by chance after allowing for effects of differences in Specimen. There is a statistically significant difference ($P = 0.011$). To isolate which group(s) differ from the others use a multiple comparison procedure.

Power of performed test with $\alpha = 0.0500$: for Specimen : 0.767

Power of performed test with $\alpha = 0.0500$: for Flap of Femur 40 or Fibula : 0.785

Least square means for Specimen :

Group	Mean	SEM
1975.000	671.578	121.616
2000.000	869.233	160.512
2017.000	736.233	160.512
2020.000	1480.000	146.526
2031.000	984.578	121.616
2008.000	916.233	160.512
2028.000	519.911	121.616

Least square means for Flap of Femur 40 or Fibula :

Group	Mean	SEM
2.000	1093.271	115.033
1.000	671.805	58.461

All Pairwise Multiple Comparison Procedures (Tukey Test):

Comparisons for factor: **Specimen**

Comparison	Diff of Means	p	q	P	P<0.050
2020.000 vs. 2028.000	960.089	7	7.130	0.008	Yes
2020.000 vs. 1975.000	808.422	7	6.004	0.024	Yes
2020.000 vs. 2017.000	743.767	7	4.840	0.075	No
2020.000 vs. 2000.000	610.767	7	3.974	0.174	Do Not Test
2020.000 vs. 2008.000	563.767	7	3.668	0.232	Do Not Test
2020.000 vs. 2031.000	495.422	7	3.679	0.230	Do Not Test
2031.000 vs. 2028.000	464.667	7	3.821	0.201	No
2031.000 vs. 1975.000	313.000	7	2.574	0.567	Do Not Test
2031.000 vs. 2017.000	248.344	7	1.744	0.865	Do Not Test
2031.000 vs. 2000.000	115.344	7	0.810	0.996	Do Not Test
2031.000 vs. 2008.000	68.344	7	0.480	1.000	Do Not Test
2008.000 vs. 2028.000	396.322	7	2.783	0.489	Do Not Test
2008.000 vs. 1975.000	244.656	7	1.718	0.872	Do Not Test
2008.000 vs. 2017.000	180.000	7	1.121	0.980	Do Not Test
2008.000 vs. 2000.000	47.000	7	0.293	1.000	Do Not Test
2000.000 vs. 2028.000	349.322	7	2.453	0.613	Do Not Test
2000.000 vs. 1975.000	197.656	7	1.388	0.946	Do Not Test
2000.000 vs. 2017.000	133.000	7	0.829	0.996	Do Not Test
2017.000 vs. 2028.000	216.322	7	1.519	0.921	Do Not Test
2017.000 vs. 1975.000	64.656	7	0.454	1.000	Do Not Test
1975.000 vs. 2028.000	151.667	7	1.247	0.967	Do Not Test

Comparisons for factor: **Flap of Femur 40 or Fibula**

Comparison	Diff of Means	p	q	P	P<0.050
2.000 vs. 1.000	421.467	2	4.619	0.010	Yes

A result of "Do Not Test" occurs for a comparison when no significant difference is found between two means that enclose that comparison. For example, if you had four means sorted in order, and found no difference between means 4 vs. 2, then you would not test 4 vs. 3 and 3 vs. 2, but still test 4 vs. 1 and 3 vs. 1 (4 vs. 3 and 3 vs. 2 are enclosed by 4 vs. 2: 4 3 2 1). Note that not testing the enclosed means is a procedural rule, and a result of Do Not Test should be treated as if there is no significant difference between the means, even though one may appear to exist.

Comparison of 40% femur flaps, taking into account the 100N safety factor, versus fibula flaps using the Least Square Difference:

Two Way Analysis of Variance

Data source: Data 1 in 40% flaps vs fibula

General Linear Model (No Interactions)

Dependent Variable: Force Minus 100 N (Femur Alone)

Normality Test: Passed (P = 0.165)

Equal Variance Test: Failed (P < 0.050)

Source of Variation	DF	SS	MS	F	P
Specimen	6	1287178.320	214529.720	4.996	0.016
Flap of Femur 40 or Fibula	1	444085.378	444085.378	10.342	0.011
Residual	9	386460.122	42940.014		
Total	16	2434995.059	152187.191		

The difference in the mean values among the different levels of Specimen is greater than would be expected by chance after allowing for effects of differences in Flap of Femur 40 or Fibula. There is a statistically significant difference (P = 0.016). To isolate which group(s) differ from the others use a multiple comparison procedure.

The difference in the mean values among the different levels of Flap of Femur 40 or Fibula is greater than would be expected by chance after allowing for effects of differences in Specimen. There is a statistically significant difference (P = 0.011). To isolate which group(s) differ from the others use a multiple comparison procedure.

Power of performed test with alpha = 0.0500: for Specimen : 0.767

Power of performed test with alpha = 0.0500: for Flap of Femur 40 or Fibula : 0.785

Least square means for Specimen :

Group	Mean	SEM
1975.000	671.578	121.616
2000.000	869.233	160.512
2017.000	736.233	160.512
2020.000	1480.000	146.526
2031.000	984.578	121.616
2008.000	916.233	160.512
2028.000	519.911	121.616

Least square means for Flap of Femur 40 or Fibula :

Group	Mean	SEM
2.000	1093.271	115.033

1.000 671.805 58.461

All Pairwise Multiple Comparison Procedures (Fisher LSD Method):

Comparisons for factor: **Specimen**

Comparison	Diff of Means	LSD(alpha=0.050)	P	Diff >= LSD
2020.000 vs. 2028.000	960.089	430.764	<0.001	Yes
2020.000 vs. 1975.000	808.422	430.764	0.002	Yes
2020.000 vs. 2017.000	743.767	491.643	0.008	Yes
2020.000 vs. 2000.000	610.767	491.643	0.020	Yes
2020.000 vs. 2008.000	563.767	491.643	0.029	Yes
2020.000 vs. 2031.000	495.422	430.764	0.029	Yes
2031.000 vs. 2028.000	464.667	389.071	0.024	Yes
2031.000 vs. 1975.000	313.000	389.071	0.102	No
2031.000 vs. 2017.000	248.344	455.556	0.249	Do Not Test
2031.000 vs. 2000.000	115.344	455.556	0.581	Do Not Test
2031.000 vs. 2008.000	68.344	455.556	0.742	Do Not Test
2008.000 vs. 2028.000	396.322	455.556	0.081	No
2008.000 vs. 1975.000	244.656	455.556	0.255	Do Not Test
2008.000 vs. 2017.000	180.000	513.505	0.448	Do Not Test
2008.000 vs. 2000.000	47.000	513.505	0.841	Do Not Test
2000.000 vs. 2028.000	349.322	455.556	0.117	Do Not Test
2000.000 vs. 1975.000	197.656	455.556	0.352	Do Not Test
2000.000 vs. 2017.000	133.000	513.505	0.572	Do Not Test
2017.000 vs. 2028.000	216.322	455.556	0.311	Do Not Test
2017.000 vs. 1975.000	64.656	455.556	0.755	Do Not Test
1975.000 vs. 2028.000	151.667	389.071	0.401	Do Not Test

Comparisons for factor: **Flap of Femur 40 or Fibula**

Comparison	Diff of Means	LSD(alpha=0.050)	P	Diff >= LSD
2.000 vs. 1.000	421.467	291.899	0.010	Yes

A result of "Do Not Test" occurs for a comparison when no significant difference is found between two means that enclose that comparison. For example, if you had four means sorted in order, and found no difference between means 4 vs. 2, then you would not test 4 vs. 3 and 3 vs. 2, but still test 4 vs. 1 and 3 vs. 1 (4 vs. 3 and 3 vs. 2 are enclosed by 4 vs. 2: 4 3 2 1). Note that not testing the enclosed means is a procedural rule, and a result of Do Not Test should be treated as if there is no significant difference between the means, even though one may appear to exist.

Table of 3-point bending results used for Sigma Plot 9.0 in comparing the 40% Femur Flaps, with a safety factor or 200N, and the Fibula Flaps:

Specimen	Flap of Femur 40 or Fibula	Force (N)	Femur Force Minus 200 N (N)
1975	Femur	810	1010-200
1975	Fibula	511	
1975	Fibula	383	
2000	Fibula	612	
2000	Fibula	705	
2017	Fibula	564	
2017	Fibula	487	
2020	Femur	1740	1940-200
2020	Fibula	1120	
2031	Femur	1310	1510-200
2031	Fibula	566	
2031	Fibula	767	
2008	Fibula	759	
2008	Fibula	652	
2028	Fibula	497	
2028	Fibula	513	
2028	Femur	239	439-200

Comparison of 40% femur flaps, taking into account the 200N safety factor, versus fibula flaps using the Tukey Test:

Two Way Analysis of Variance

Data source: Data 1 in Notebook 2

General Linear Model (No Interactions)

Dependent Variable: Force Minus 200 N (Femur Alone)

Normality Test: Passed (P = 0.165)

Equal Variance Test: Failed (P < 0.050)

Source of Variation	DF	SS	MS	F	P
Specimen	6	1287178.320	214529.720	4.996	0.016
Flap of Femur 40 or Fibula	1	258352.044	258352.044	6.017	0.037
Residual	9	386460.122	42940.014		
Total	16	2160371.529	135023.221		

The difference in the mean values among the different levels of Specimen is greater than would be expected by chance after allowing for effects of differences in Flap of Femur 40 or Fibula. There is a statistically significant difference (P = 0.016). To isolate which group(s) differ from the others use a multiple comparison procedure.

The difference in the mean values among the different levels of Flap of Femur 40 or Fibula is greater than would be expected by chance after allowing for effects of differences in Specimen. There is a statistically significant difference ($P = 0.037$). To isolate which group(s) differ from the others use a multiple comparison procedure.

Power of performed test with $\alpha = 0.0500$: for Specimen : 0.767

Power of performed test with $\alpha = 0.0500$: for Flap of Femur 40 or Fibula : 0.511

Least square means for Specimen :

Group	Mean	SEM
1975.000	621.578	121.616
2000.000	819.233	160.512
2017.000	686.233	160.512
2020.000	1430.000	146.526
2031.000	934.578	121.616
2008.000	866.233	160.512
2028.000	469.911	121.616

Least square means for Flap of Femur 40 or Fibula :

Group	Mean	SEM
2.000	993.271	115.033
1.000	671.805	58.461

All Pairwise Multiple Comparison Procedures (Tukey Test):

Comparisons for factor: **Specimen**

Comparison	Diff of Means	p	q	P	P<0.050
2020.000 vs. 2028.000	960.089	7	7.130	0.008	Yes
2020.000 vs. 1975.000	808.422	7	6.004	0.024	Yes
2020.000 vs. 2017.000	743.767	7	4.840	0.075	No
2020.000 vs. 2000.000	610.767	7	3.974	0.174	Do Not Test
2020.000 vs. 2008.000	563.767	7	3.668	0.232	Do Not Test
2020.000 vs. 2031.000	495.422	7	3.679	0.230	Do Not Test
2031.000 vs. 2028.000	464.667	7	3.821	0.201	No
2031.000 vs. 1975.000	313.000	7	2.574	0.567	Do Not Test
2031.000 vs. 2017.000	248.344	7	1.744	0.865	Do Not Test
2031.000 vs. 2000.000	115.344	7	0.810	0.996	Do Not Test
2031.000 vs. 2008.000	68.344	7	0.480	1.000	Do Not Test
2008.000 vs. 2028.000	396.322	7	2.783	0.489	Do Not Test
2008.000 vs. 1975.000	244.656	7	1.718	0.872	Do Not Test
2008.000 vs. 2017.000	180.000	7	1.121	0.980	Do Not Test
2008.000 vs. 2000.000	47.000	7	0.293	1.000	Do Not Test
2000.000 vs. 2028.000	349.322	7	2.453	0.613	Do Not Test
2000.000 vs. 1975.000	197.656	7	1.388	0.946	Do Not Test
2000.000 vs. 2017.000	133.000	7	0.829	0.996	Do Not Test
2017.000 vs. 2028.000	216.322	7	1.519	0.921	Do Not Test
2017.000 vs. 1975.000	64.656	7	0.454	1.000	Do Not Test
1975.000 vs. 2028.000	151.667	7	1.247	0.967	Do Not Test

Comparisons for factor: **Flap of Femur 40 or Fibula**

Comparison	Diff of Means	p	q	P	P<0.050
2.000 vs. 1.000	321.467	2	3.523	0.035	Yes

A result of "Do Not Test" occurs for a comparison when no significant difference is found between two means that enclose that comparison. For example, if you had four means sorted in order, and found no difference between means 4 vs. 2, then you would not test 4 vs. 3 and 3 vs. 2, but still test 4 vs. 1 and 3 vs. 1 (4 vs. 3 and 3 vs. 2 are enclosed by 4 vs. 2: 4 3 2 1). Note that not testing the enclosed means is a procedural rule, and a result of Do Not Test should be treated as if there is no significant difference between the means, even though one may appear to exist.

Comparison of 40% femur flaps, taking into account the 200N safety factor, versus fibula flaps using the Least Square Difference:

Two Way Analysis of Variance

Data source: Data 1 in Notebook 2

General Linear Model (No Interactions)

Dependent Variable: Force Minus 200 N (Femur Alone)

Normality Test: Passed (P = 0.165)

Equal Variance Test: Failed (P < 0.050)

Source of Variation	DF	SS	MS	F	P
Specimen	6	1287178.320	214529.720	4.996	0.016
Flap of Femur 40 or Fibula	1	258352.044	258352.044	6.017	0.037
Residual	9	386460.122	42940.014		
Total	16	2160371.529	135023.221		

The difference in the mean values among the different levels of Specimen is greater than would be expected by chance after allowing for effects of differences in Flap of Femur 40 or Fibula. There is a statistically significant difference (P = 0.016). To isolate which group(s) differ from the others use a multiple comparison procedure.

The difference in the mean values among the different levels of Flap of Femur 40 or Fibula is greater than would be expected by chance after allowing for effects of differences in Specimen. There is a statistically significant difference (P = 0.037). To isolate which group(s) differ from the others use a multiple comparison procedure.

Power of performed test with alpha = 0.0500: for Specimen : 0.767

Power of performed test with alpha = 0.0500: for Flap of Femur 40 or Fibula : 0.511

Least square means for Specimen :

Group	Mean	SEM
1975.000	621.578	121.616
2000.000	819.233	160.512
2017.000	686.233	160.512
2020.000	1430.000	146.526
2031.000	934.578	121.616
2008.000	866.233	160.512
2028.000	469.911	121.616

Least square means for Flap of Femur 40 or Fibula :

Group	Mean	SEM
2.000	993.271	115.033

1.000 671.805 58.461

All Pairwise Multiple Comparison Procedures (Fisher LSD Method):

Comparisons for factor: **Specimen**

Comparison	Diff of Means	LSD(alpha=0.050)	P	Diff >= LSD
2020.000 vs. 2028.000	960.089	430.764	<0.001	Yes
2020.000 vs. 1975.000	808.422	430.764	0.002	Yes
2020.000 vs. 2017.000	743.767	491.643	0.008	Yes
2020.000 vs. 2000.000	610.767	491.643	0.020	Yes
2020.000 vs. 2008.000	563.767	491.643	0.029	Yes
2020.000 vs. 2031.000	495.422	430.764	0.029	Yes
2031.000 vs. 2028.000	464.667	389.071	0.024	Yes
2031.000 vs. 1975.000	313.000	389.071	0.102	No
2031.000 vs. 2017.000	248.344	455.556	0.249	Do Not Test
2031.000 vs. 2000.000	115.344	455.556	0.581	Do Not Test
2031.000 vs. 2008.000	68.344	455.556	0.742	Do Not Test
2008.000 vs. 2028.000	396.322	455.556	0.081	No
2008.000 vs. 1975.000	244.656	455.556	0.255	Do Not Test
2008.000 vs. 2017.000	180.000	513.505	0.448	Do Not Test
2008.000 vs. 2000.000	47.000	513.505	0.841	Do Not Test
2000.000 vs. 2028.000	349.322	455.556	0.117	Do Not Test
2000.000 vs. 1975.000	197.656	455.556	0.352	Do Not Test
2000.000 vs. 2017.000	133.000	513.505	0.572	Do Not Test
2017.000 vs. 2028.000	216.322	455.556	0.311	Do Not Test
2017.000 vs. 1975.000	64.656	455.556	0.755	Do Not Test
1975.000 vs. 2028.000	151.667	389.071	0.401	Do Not Test

Comparisons for factor: **Flap of Femur 40 or Fibula**

Comparison	Diff of Means	LSD(alpha=0.050)	P	Diff >= LSD
2.000 vs. 1.000	321.467	291.899	0.034	Yes

A result of "Do Not Test" occurs for a comparison when no significant difference is found between two means that enclose that comparison. For example, if you had four means sorted in order, and found no difference between means 4 vs. 2, then you would not test 4 vs. 3 and 3 vs. 2, but still test 4 vs. 1 and 3 vs. 1 (4 vs. 3 and 3 vs. 2 are enclosed by 4 vs. 2: 4 3 2 1). Note that not testing the enclosed means is a procedural rule, and a result of Do Not Test should be treated as if there is no significant difference between the means, even though one may appear to exist.

Table of 3-point bending results used for Sigma Plot 9.0 in comparing the 40% Femur Flaps, with a safety factor or 225N, and the Fibula Flaps:

Specimen	Flap of Femur 40 or Fibula	Force (N)	Femur Force Minus 225 N (N)
1975	Femur	785	1010-225
1975	Fibula	511	
1975	Fibula	383	
2000	Fibula	612	
2000	Fibula	705	
2017	Fibula	564	
2017	Fibula	487	
2020	Femur	1715	1940-225
2020	Fibula	1120	
2031	Femur	1285	1510-225
2031	Fibula	566	
2031	Fibula	767	
2008	Fibula	759	
2008	Fibula	652	
2028	Fibula	497	
2028	Fibula	513	
2028	Femur	214	439-225

Comparison of 40% femur flaps, taking into account the 225N safety factor, versus fibula flaps using the Tukey Test:

Two Way Analysis of Variance

Data source: Data 1 in Notebook 3

General Linear Model (No Interactions)

Dependent Variable: Force Minus 225 N (Femur Alone)

Normality Test: Passed (P = 0.165)

Equal Variance Test: Failed (P < 0.050)

Source of Variation	DF	SS	MS	F	P
Specimen	6	1287178.320	214529.720	4.996	0.016
Flap of Femur 40 or Fibula	1	219731.211	219731.211	5.117	0.050
Residual	9	386460.122	42940.014		
Total	16	2101274.471	131329.654		

The difference in the mean values among the different levels of Specimen is greater than would be expected by chance after allowing for effects of differences in Flap of Femur 40 or Fibula. There is a statistically significant difference (P = 0.016). To isolate which group(s) differ from the others use a multiple comparison procedure.

The difference in the mean values among the different levels of Flap of Femur 40 or Fibula is not great enough to exclude the possibility that the difference is just due to random sampling variability after allowing for the effects of differences in Specimen. There is not a statistically significant difference (P = 0.050).

Power of performed test with alpha = 0.0500: for Specimen : 0.767

Power of performed test with alpha = 0.0500: for Flap of Femur 40 or Fibula : 0.434

Least square means for Specimen :

Group	Mean	SEM
1975.000	609.078	121.616
2000.000	806.733	160.512
2017.000	673.733	160.512
2020.000	1417.500	146.526
2031.000	922.078	121.616
2008.000	853.733	160.512
2028.000	457.411	121.616

Least square means for Flap of Femur 40 or Fibula :

Group	Mean	SEM
2.000	968.271	115.033
1.000	671.805	58.461

All Pairwise Multiple Comparison Procedures (Tukey Test):

Comparisons for factor: **Specimen**

Comparison	Diff of Means	p	q	P	P<0.050
2020.000 vs. 2028.000	960.089	7	7.130	0.008	Yes
2020.000 vs. 1975.000	808.422	7	6.004	0.024	Yes
2020.000 vs. 2017.000	743.767	7	4.840	0.075	No
2020.000 vs. 2000.000	610.767	7	3.974	0.174	Do Not Test
2020.000 vs. 2008.000	563.767	7	3.668	0.232	Do Not Test
2020.000 vs. 2031.000	495.422	7	3.679	0.230	Do Not Test
2031.000 vs. 2028.000	464.667	7	3.821	0.201	No
2031.000 vs. 1975.000	313.000	7	2.574	0.567	Do Not Test
2031.000 vs. 2017.000	248.344	7	1.744	0.865	Do Not Test
2031.000 vs. 2000.000	115.344	7	0.810	0.996	Do Not Test
2031.000 vs. 2008.000	68.344	7	0.480	1.000	Do Not Test
2008.000 vs. 2028.000	396.322	7	2.783	0.489	Do Not Test
2008.000 vs. 1975.000	244.656	7	1.718	0.872	Do Not Test
2008.000 vs. 2017.000	180.000	7	1.121	0.980	Do Not Test
2008.000 vs. 2000.000	47.000	7	0.293	1.000	Do Not Test
2000.000 vs. 2028.000	349.322	7	2.453	0.613	Do Not Test
2000.000 vs. 1975.000	197.656	7	1.388	0.946	Do Not Test
2000.000 vs. 2017.000	133.000	7	0.829	0.996	Do Not Test
2017.000 vs. 2028.000	216.322	7	1.519	0.921	Do Not Test
2017.000 vs. 1975.000	64.656	7	0.454	1.000	Do Not Test
1975.000 vs. 2028.000	151.667	7	1.247	0.967	Do Not Test

Comparisons for factor: **Flap of Femur 40 or Fibula**

Comparison	Diff of Means	p	q	P	P<0.050
2.000 vs. 1.000	296.467	2	3.249	0.047	Yes

A result of "Do Not Test" occurs for a comparison when no significant difference is found between two means that enclose that comparison. For example, if you had four means sorted in order, and found no difference between means 4 vs. 2, then you would not test 4 vs. 3 and 3 vs. 2, but still test 4 vs. 1 and 3 vs. 1 (4 vs. 3 and 3 vs. 2 are enclosed by 4 vs. 2: 4 3 2 1). Note that not testing the enclosed means is a procedural rule, and a result of Do Not Test should be treated as if there is no significant difference between the means, even though one may appear to exist.

Comparison of 40% femur flaps, taking into account the 225N safety factor, versus fibula flaps using the Least Square Difference:

Two Way Analysis of Variance

Data source: Data 1 in Notebook 3

General Linear Model (No Interactions)

Dependent Variable: Force Minus 225 N (Femur Alone)

Normality Test: Passed (P = 0.165)

Equal Variance Test: Failed (P < 0.050)

Source of Variation	DF	SS	MS	F	P
Specimen	6	1287178.320	214529.720	4.996	0.016
Flap of Femur 40 or Fibula	1	219731.211	219731.211	5.117	0.050
Residual	9	386460.122	42940.014		
Total	16	2101274.471	131329.654		

The difference in the mean values among the different levels of Specimen is greater than would be expected by chance after allowing for effects of differences in Flap of Femur 40 or Fibula. There is a statistically significant difference (P = 0.016). To isolate which group(s) differ from the others use a multiple comparison procedure.

The difference in the mean values among the different levels of Flap of Femur 40 or Fibula is not great enough to exclude the possibility that the difference is just due to random sampling variability after allowing for the effects of differences in Specimen. There is not a statistically significant difference (P = 0.050).

Power of performed test with alpha = 0.0500: for Specimen : 0.767

Power of performed test with alpha = 0.0500: for Flap of Femur 40 or Fibula : 0.434

Least square means for Specimen :

Group	Mean	SEM
1975.000	609.078	121.616
2000.000	806.733	160.512
2017.000	673.733	160.512
2020.000	1417.500	146.526
2031.000	922.078	121.616
2008.000	853.733	160.512
2028.000	457.411	121.616

Least square means for Flap of Femur 40 or Fibula :

Group	Mean	SEM
2.000	968.271	115.033

1.000 671.805 58.461

All Pairwise Multiple Comparison Procedures (Fisher LSD Method):

Comparisons for factor: **Specimen**

Comparison	Diff of Means	LSD(alpha=0.050)	P	Diff >= LSD
2020.000 vs. 2028.000	960.089	430.764	<0.001	Yes
2020.000 vs. 1975.000	808.422	430.764	0.002	Yes
2020.000 vs. 2017.000	743.767	491.643	0.008	Yes
2020.000 vs. 2000.000	610.767	491.643	0.020	Yes
2020.000 vs. 2008.000	563.767	491.643	0.029	Yes
2020.000 vs. 2031.000	495.422	430.764	0.029	Yes
2031.000 vs. 2028.000	464.667	389.071	0.024	Yes
2031.000 vs. 1975.000	313.000	389.071	0.102	No
2031.000 vs. 2017.000	248.344	455.556	0.249	Do Not Test
2031.000 vs. 2000.000	115.344	455.556	0.581	Do Not Test
2031.000 vs. 2008.000	68.344	455.556	0.742	Do Not Test
2008.000 vs. 2028.000	396.322	455.556	0.081	No
2008.000 vs. 1975.000	244.656	455.556	0.255	Do Not Test
2008.000 vs. 2017.000	180.000	513.505	0.448	Do Not Test
2008.000 vs. 2000.000	47.000	513.505	0.841	Do Not Test
2000.000 vs. 2028.000	349.322	455.556	0.117	Do Not Test
2000.000 vs. 1975.000	197.656	455.556	0.352	Do Not Test
2000.000 vs. 2017.000	133.000	513.505	0.572	Do Not Test
2017.000 vs. 2028.000	216.322	455.556	0.311	Do Not Test
2017.000 vs. 1975.000	64.656	455.556	0.755	Do Not Test
1975.000 vs. 2028.000	151.667	389.071	0.401	Do Not Test

Comparisons for factor: **Flap of Femur 40 or Fibula**

Comparison	Diff of Means	LSD(alpha=0.050)	P	Diff >= LSD
2.000 vs. 1.000	296.467	291.899	0.047	Yes

A result of "Do Not Test" occurs for a comparison when no significant difference is found between two means that enclose that comparison. For example, if you had four means sorted in order, and found no difference between means 4 vs. 2, then you would not test 4 vs. 3 and 3 vs. 2, but still test 4 vs. 1 and 3 vs. 1 (4 vs. 3 and 3 vs. 2 are enclosed by 4 vs. 2: 4 3 2 1). Note that not testing the enclosed means is a procedural rule, and a result of Do Not Test should be treated as if there is no significant difference between the means, even though one may appear to exist.

Table of 3-point bending results used for Sigma Plot 9.0 in comparing the 40% Femur Flaps, with a safety factor or 250N, and the Fibula Flaps:

Specimen	Flap of Femur 40 or Fibula	Force (N)	Femur Force Minus 250 N (N)
1975	Femur	760	1010-250
1975	Fibula	511	
1975	Fibula	383	
2000	Fibula	612	
2000	Fibula	705	
2017	Fibula	564	
2017	Fibula	487	
2020	Femur	1690	1940-250
2020	Fibula	1120	
2031	Femur	1260	1510-250
2031	Fibula	566	
2031	Fibula	767	
2008	Fibula	759	
2008	Fibula	652	
2028	Fibula	497	
2028	Fibula	513	
2028	Femur	189	439-250

Comparison of 40% femur flaps, taking into account the 250N safety factor, versus fibula flaps using the Tukey Test:

Two Way Analysis of Variance

Data source: Data 1 in Notebook 2

General Linear Model (No Interactions)

Dependent Variable: Force Minus 250 N (Femur Alone)

Normality Test: Passed (P = 0.165)

Equal Variance Test: Failed (P < 0.050)

Source of Variation	DF	SS	MS	F	P
Specimen	6	1287178.320	214529.720	4.996	0.016
Flap of Femur 40 or Fibula	1	184235.378	184235.378	4.291	0.068
Residual	9	386460.122	42940.014		
Total	16	2046000.941	127875.059		

The difference in the mean values among the different levels of Specimen is greater than would be expected by chance after allowing for effects of differences in Flap of Femur 40 or Fibula. There is a statistically significant difference (P = 0.016). To isolate which group(s) differ from the others use a multiple comparison procedure.

The difference in the mean values among the different levels of Flap of Femur 40 or Fibula is not great enough to exclude the possibility that the difference is just due to random sampling variability after allowing for the effects of differences in Specimen. There is not a statistically significant difference (P = 0.068).

Power of performed test with alpha = 0.0500: for Specimen : 0.767

Power of performed test with alpha = 0.0500: for Flap of Femur 40 or Fibula : 0.358

Least square means for Specimen :

Group	Mean	SEM
1975.000	596.578	121.616
2000.000	794.233	160.512
2017.000	661.233	160.512
2020.000	1405.000	146.526
2031.000	909.578	121.616
2008.000	841.233	160.512
2028.000	444.911	121.616

Least square means for Flap of Femur 40 or Fibula :

Group	Mean	SEM
2.000	943.271	115.033
1.000	671.805	58.461

All Pairwise Multiple Comparison Procedures (Tukey Test):

Comparisons for factor: **Specimen**

Comparison	Diff of Means	p	q	P	P<0.050
2020.000 vs. 2028.000	960.089	7	7.130	0.008	Yes
2020.000 vs. 1975.000	808.422	7	6.004	0.024	Yes
2020.000 vs. 2017.000	743.767	7	4.840	0.075	No
2020.000 vs. 2000.000	610.767	7	3.974	0.174	Do Not Test
2020.000 vs. 2008.000	563.767	7	3.668	0.232	Do Not Test
2020.000 vs. 2031.000	495.422	7	3.679	0.230	Do Not Test
2031.000 vs. 2028.000	464.667	7	3.821	0.201	No
2031.000 vs. 1975.000	313.000	7	2.574	0.567	Do Not Test
2031.000 vs. 2017.000	248.344	7	1.744	0.865	Do Not Test
2031.000 vs. 2000.000	115.344	7	0.810	0.996	Do Not Test
2031.000 vs. 2008.000	68.344	7	0.480	1.000	Do Not Test
2008.000 vs. 2028.000	396.322	7	2.783	0.489	Do Not Test
2008.000 vs. 1975.000	244.656	7	1.718	0.872	Do Not Test
2008.000 vs. 2017.000	180.000	7	1.121	0.980	Do Not Test
2008.000 vs. 2000.000	47.000	7	0.293	1.000	Do Not Test
2000.000 vs. 2028.000	349.322	7	2.453	0.613	Do Not Test
2000.000 vs. 1975.000	197.656	7	1.388	0.946	Do Not Test
2000.000 vs. 2017.000	133.000	7	0.829	0.996	Do Not Test
2017.000 vs. 2028.000	216.322	7	1.519	0.921	Do Not Test
2017.000 vs. 1975.000	64.656	7	0.454	1.000	Do Not Test
1975.000 vs. 2028.000	151.667	7	1.247	0.967	Do Not Test

Comparisons for factor: **Flap of Femur 40 or Fibula**

Comparison	Diff of Means	p	q	P	P<0.050
2.000 vs. 1.000	271.467	2	2.975	0.065	No

A result of "Do Not Test" occurs for a comparison when no significant difference is found between two means that enclose that comparison. For example, if you had four means sorted in order, and found no difference between means 4 vs. 2, then you would not test 4 vs. 3 and 3 vs. 2, but still test 4 vs. 1 and 3 vs. 1 (4 vs. 3 and 3 vs. 2 are enclosed by 4 vs. 2: 4 3 2 1). Note that not testing the enclosed means is a procedural rule, and a result of Do Not Test should be treated as if there is no significant difference between the means, even though one may appear to exist.

Comparison of 40% femur flaps, taking into account the 250N safety factor, versus fibula flaps using the Least Square Difference:

Two Way Analysis of Variance

Data source: Data 1 in Notebook 2

General Linear Model (No Interactions)

Dependent Variable: Force Minus 250 N (Femur Alone)

Normality Test: Passed (P = 0.165)

Equal Variance Test: Failed (P < 0.050)

Source of Variation	DF	SS	MS	F	P
Specimen	6	1287178.320	214529.720	4.996	0.016
Flap of Femur 40 or Fibula	1	184235.378	184235.378	4.291	0.068
Residual	9	386460.122	42940.014		
Total	16	2046000.941	127875.059		

The difference in the mean values among the different levels of Specimen is greater than would be expected by chance after allowing for effects of differences in Flap of Femur 40 or Fibula. There is a statistically significant difference (P = 0.016). To isolate which group(s) differ from the others use a multiple comparison procedure.

The difference in the mean values among the different levels of Flap of Femur 40 or Fibula is not great enough to exclude the possibility that the difference is just due to random sampling variability after allowing for the effects of differences in Specimen. There is not a statistically significant difference (P = 0.068).

Power of performed test with alpha = 0.0500: for Specimen : 0.767

Power of performed test with alpha = 0.0500: for Flap of Femur 40 or Fibula : 0.358

Least square means for Specimen :

Group	Mean	SEM
1975.000	596.578	121.616
2000.000	794.233	160.512
2017.000	661.233	160.512
2020.000	1405.000	146.526
2031.000	909.578	121.616
2008.000	841.233	160.512
2028.000	444.911	121.616

Least square means for Flap of Femur 40 or Fibula :

Group	Mean	SEM
2.000	943.271	115.033
1.000	671.805	58.461

All Pairwise Multiple Comparison Procedures (Fisher LSD Method):

Comparisons for factor: **Specimen**

Comparison	Diff of Means	LSD(alpha=0.050)	P	Diff >= LSD
2020.000 vs. 2028.000	960.089	430.764	<0.001	Yes
2020.000 vs. 1975.000	808.422	430.764	0.002	Yes
2020.000 vs. 2017.000	743.767	491.643	0.008	Yes
2020.000 vs. 2000.000	610.767	491.643	0.020	Yes
2020.000 vs. 2008.000	563.767	491.643	0.029	Yes
2020.000 vs. 2031.000	495.422	430.764	0.029	Yes
2031.000 vs. 2028.000	464.667	389.071	0.024	Yes
2031.000 vs. 1975.000	313.000	389.071	0.102	No
2031.000 vs. 2017.000	248.344	455.556	0.249	Do Not Test
2031.000 vs. 2000.000	115.344	455.556	0.581	Do Not Test
2031.000 vs. 2008.000	68.344	455.556	0.742	Do Not Test
2008.000 vs. 2028.000	396.322	455.556	0.081	No
2008.000 vs. 1975.000	244.656	455.556	0.255	Do Not Test
2008.000 vs. 2017.000	180.000	513.505	0.448	Do Not Test
2008.000 vs. 2000.000	47.000	513.505	0.841	Do Not Test
2000.000 vs. 2028.000	349.322	455.556	0.117	Do Not Test
2000.000 vs. 1975.000	197.656	455.556	0.352	Do Not Test
2000.000 vs. 2017.000	133.000	513.505	0.572	Do Not Test
2017.000 vs. 2028.000	216.322	455.556	0.311	Do Not Test
2017.000 vs. 1975.000	64.656	455.556	0.755	Do Not Test
1975.000 vs. 2028.000	151.667	389.071	0.401	Do Not Test

Comparisons for factor: **Flap of Femur 40 or Fibula**

Comparison	Diff of Means	LSD(alpha=0.050)	P	Diff >= LSD
2.000 vs. 1.000	271.467	291.899	0.065	No

A result of "Do Not Test" occurs for a comparison when no significant difference is found between two means that enclose that comparison. For example, if you had four means sorted in order, and found no difference between means 4 vs. 2, then you would not test 4 vs. 3 and 3 vs. 2, but still test 4 vs. 1 and 3 vs. 1 (4 vs. 3 and 3 vs. 2 are enclosed by 4 vs. 2: 4 3 2 1). Note that not testing the enclosed means is a procedural rule, and a result of Do Not Test should be treated as if there is no significant difference between the means, even though one may appear to exist.

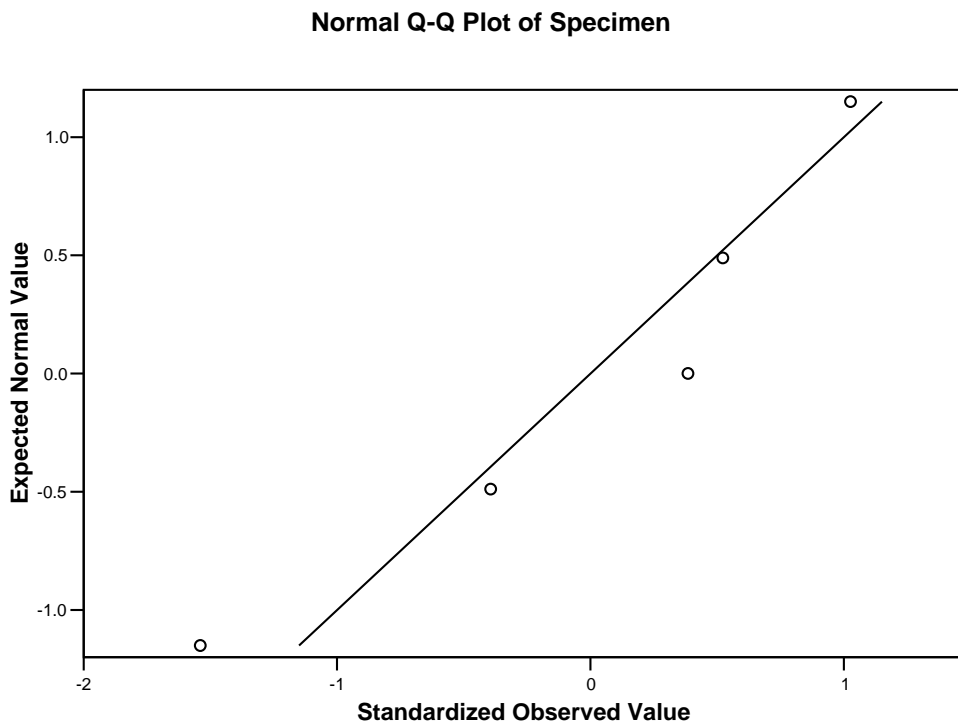
35% Harvested Femur Tested for Outliers:

PPlot

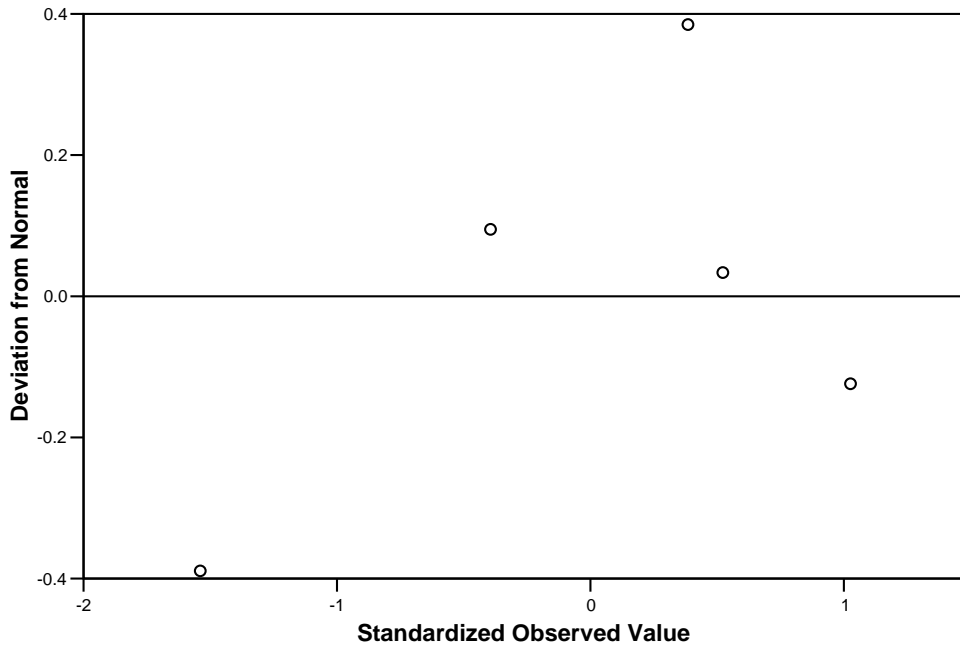
MODEL: MOD_5.
Distribution tested: Normal
Proportion estimation formula used: Tukey's
Rank assigned to ties: Mean

—

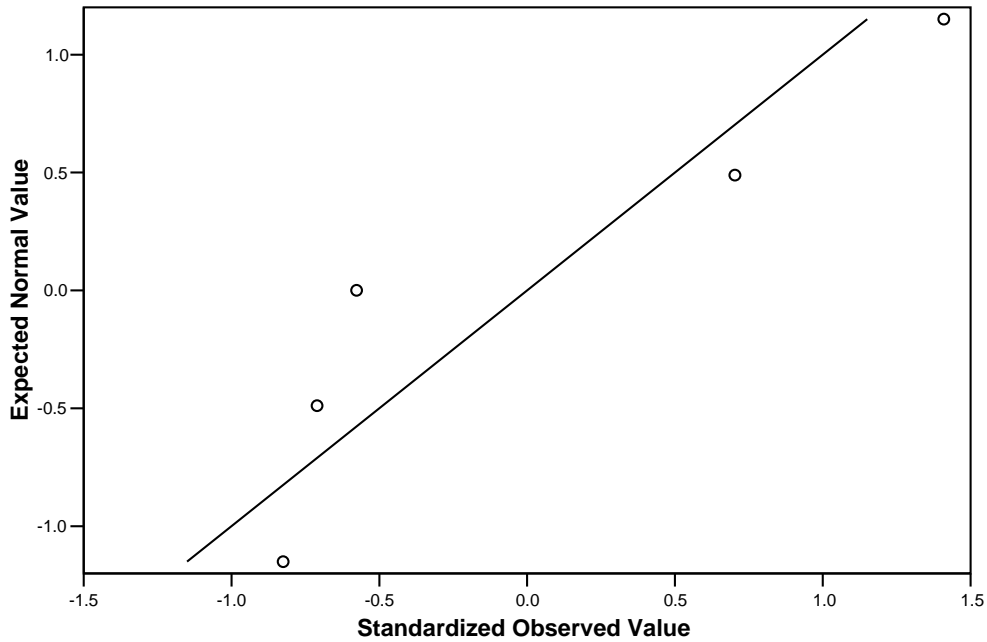
For variable Specimen ...
Normal distribution parameters estimated: location = 0 and scale = 1
For variable FailureForceN ...
Normal distribution parameters estimated: location = 0 and scale = 1



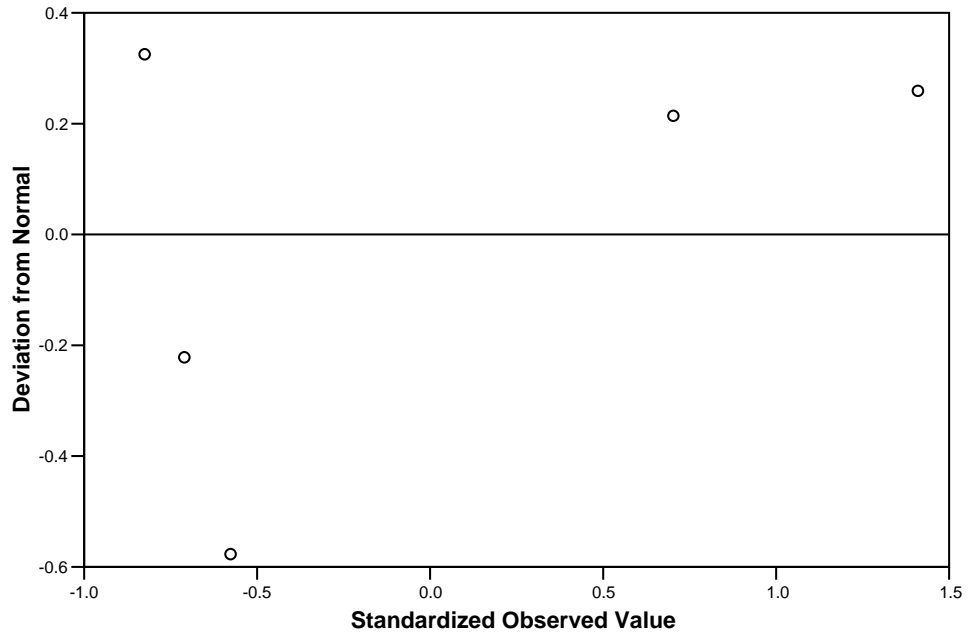
Detrended Normal Q-Q Plot of Specimen



Normal Q-Q Plot of Failure Force (N)



Detrended Normal Q-Q Plot of Failure Force (N)



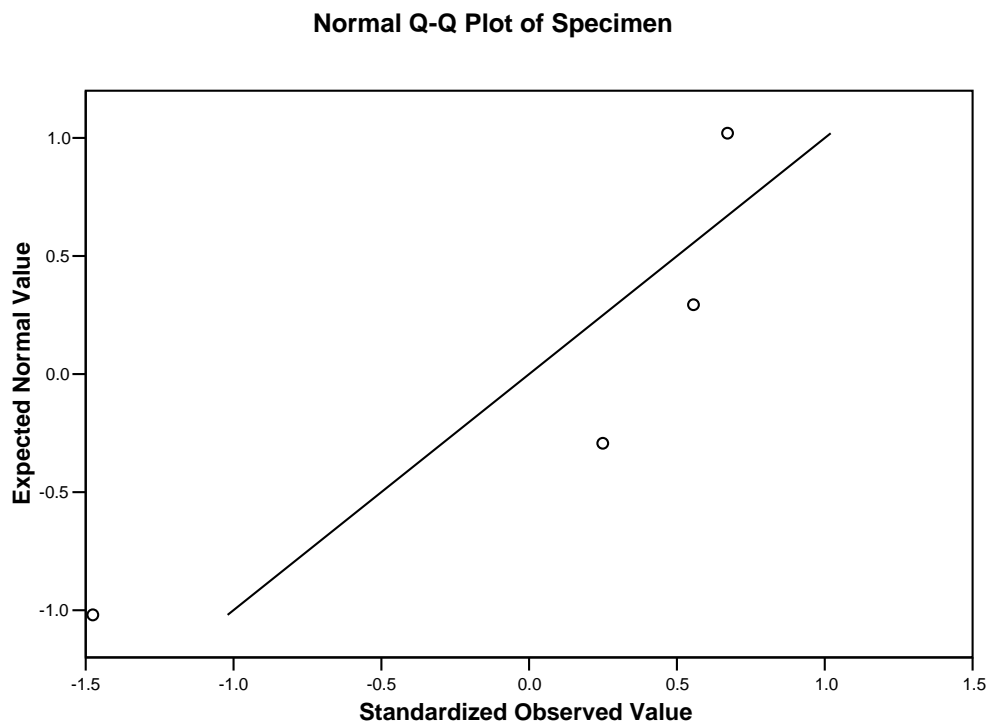
40% Harvested Femur Tested for Outliers:

PPlot

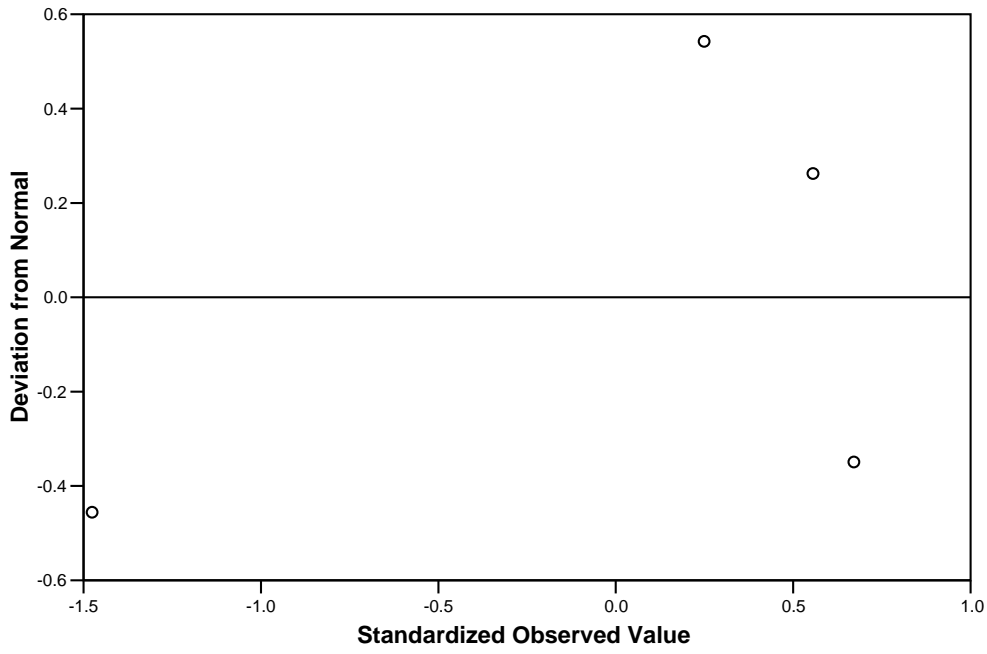
MODEL: MOD_6.
Distribution tested: Normal
Proportion estimation formula used: Tukey's
Rank assigned to ties: Mean

—

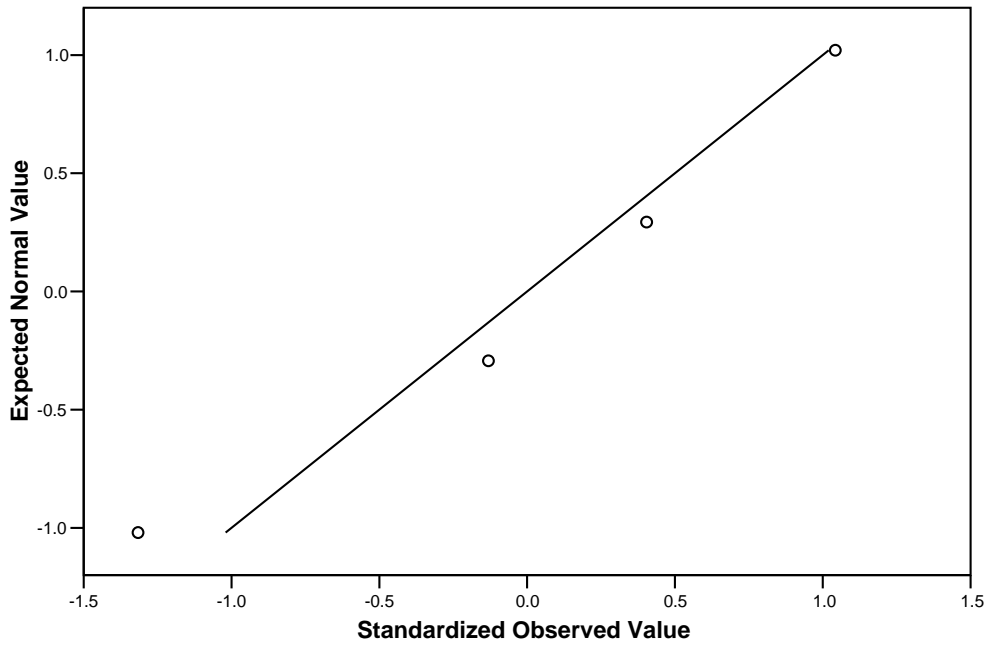
For variable Specimen ...
Normal distribution parameters estimated: location = 0 and scale = 1
For variable FailureForceN ...
Normal distribution parameters estimated: location = 0 and scale = 1



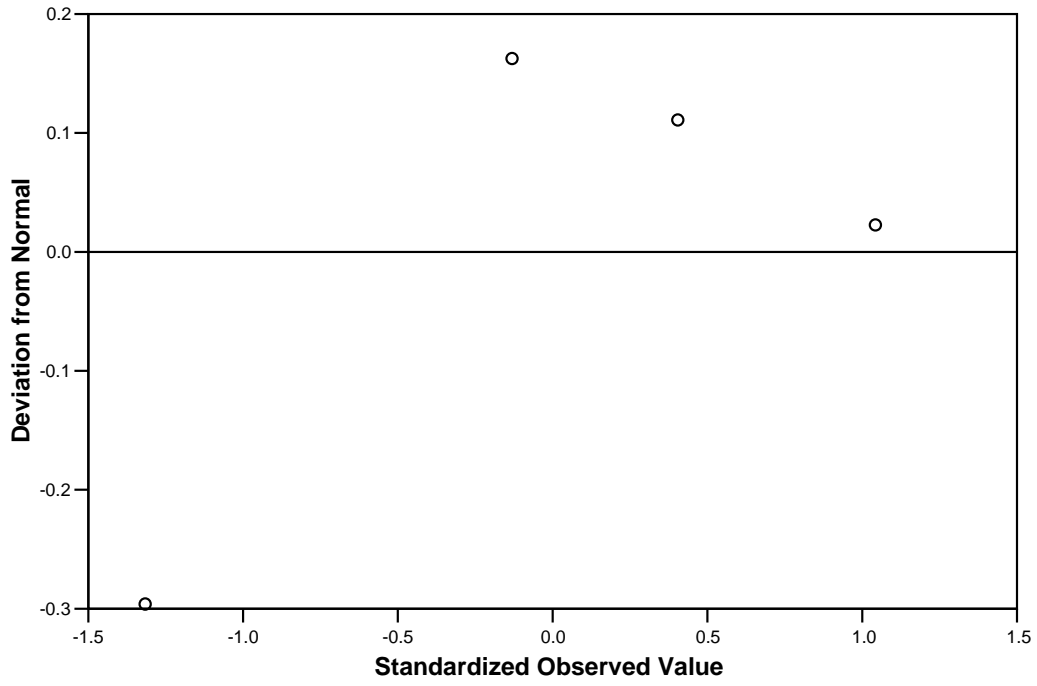
Detrended Normal Q-Q Plot of Specimen



Normal Q-Q Plot of Failure Force (N)



Detrended Normal Q-Q Plot of Failure Force (N)



Whole Femur Tested for Outliers:

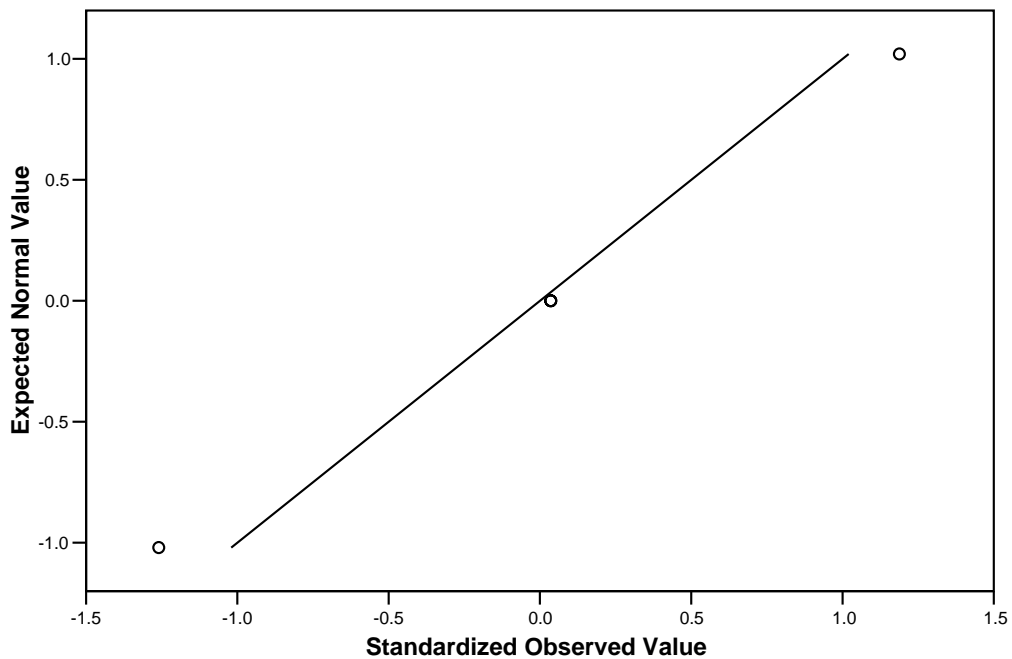
PPlot

MODEL: MOD_7.
Distribution tested: Normal
Proportion estimation formula used: Tukey's
Rank assigned to ties: Mean

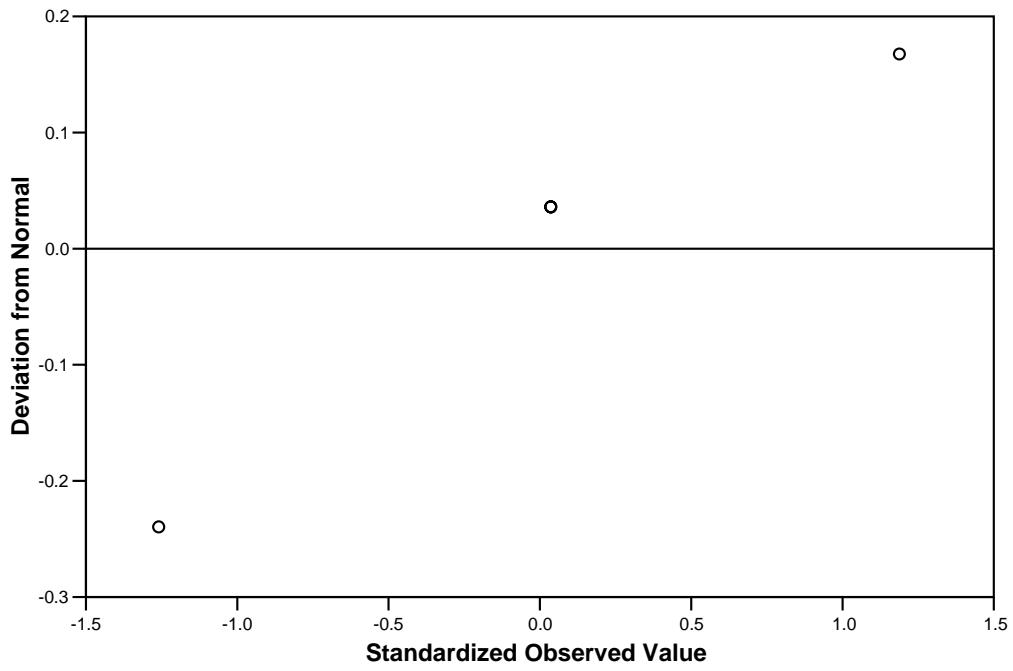
—

For variable Specimen ...
Normal distribution parameters estimated: location = 0 and scale = 1
For variable FailureForceN ...
Normal distribution parameters estimated: location = 0 and scale = 1

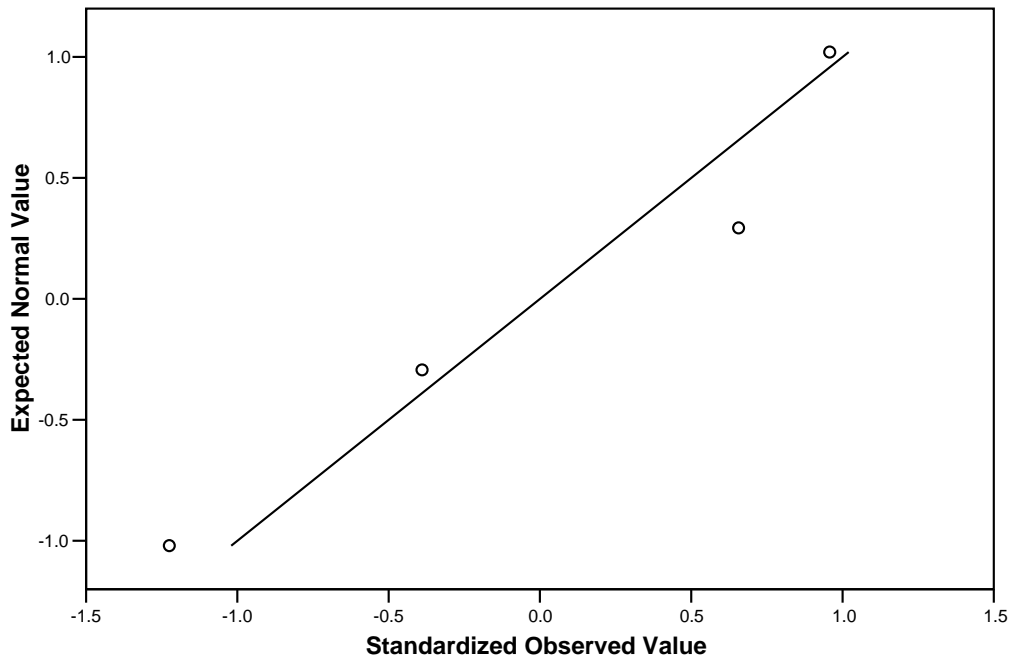
Normal Q-Q Plot of Specimen



Detrended Normal Q-Q Plot of Specimen



Normal Q-Q Plot of Failure Force (N)



Detrended Normal Q-Q Plot of Failure Force (N)

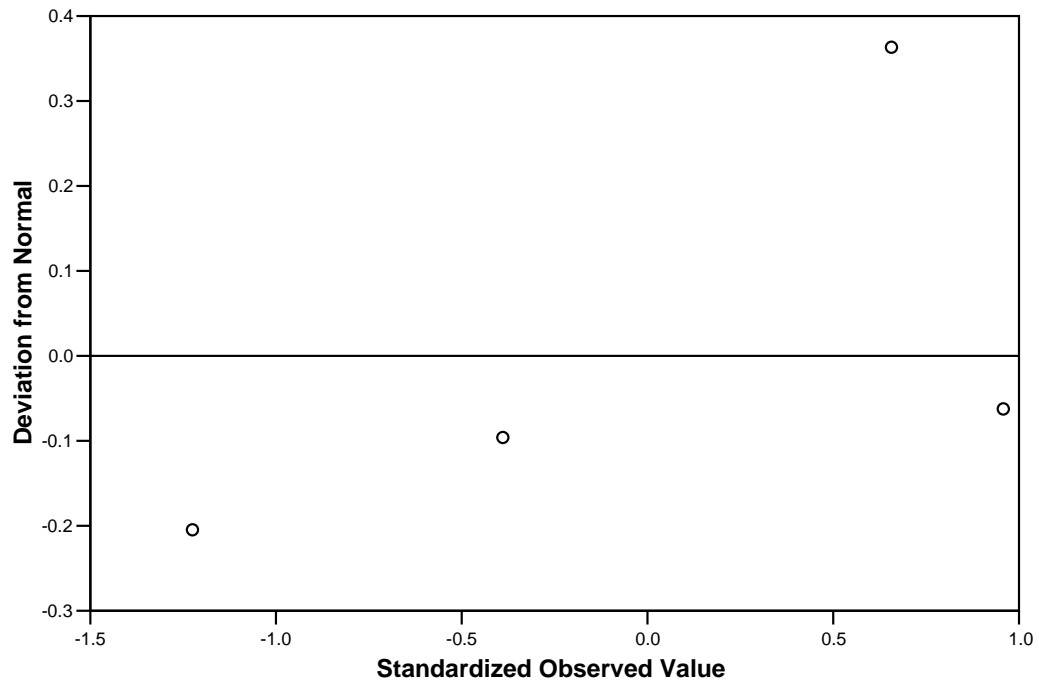


Table of compression testing results used for Sigma Plot 9.0 in comparing the 35% Harvested Femurs and the Whole Femurs:

Specimen	Whole/35% Harvested	Failure Force (N)
2017	Harvested	2740
2017	Whole	3460
2000	Harvested	2860
2000	Whole	2350
1975	Harvested	4340
2031	Harvested	3000
2020	Harvested	5080
2009	Whole	4850
2009	Whole	5250

Comparison of the 35% harvested femur versus the whole femur using a one-tailed t-test:

Two Way Analysis of Variance

Data source: Data 1 in 35Harvested vs Whole femur

General Linear Model (No Interactions)

Dependent Variable: Failure Force (N)

Normality Test: Passed (P = 0.080)

Equal Variance Test: Passed (P = 1.000)

Source of Variation	DF	SS	MS	F	P
Specimen	5	9223970.000	1844794.000	8.052	0.114
Whole/Harvested	1	11025.000	11025.000	0.0481	0.847
Residual	2	458225.000	229112.500		
Total	8	9992200.000	1249025.000		

The difference in the mean values among the different levels of Specimen is not great enough to exclude the possibility that the difference is just due to random sampling variability after allowing for the effects of differences in Whole/Harvested. There is not a statistically significant difference (P = 0.114).

The difference in the mean values among the different levels of Whole/Harvested is not great enough to exclude the possibility that the difference is just due to random sampling variability after allowing for the effects of differences in Specimen. There is not a statistically significant difference (P = 0.847).

Power of performed test with alpha = 0.0500: for Specimen : 0.331

Power of performed test with alpha = 0.0500: for Whole/Harvested : 0.0578

Least square means for Specimen :

Group	Mean	SEM
2017.000	3100.000	338.462
2000.000	2605.000	338.462
1975.000	4392.500	535.155
2031.000	3052.500	535.155
2020.000	5132.500	535.155
2009.000	4997.500	414.529

Least square means for Whole/Harvested :

Group	Mean	SEM
2.000	3827.500	232.585
1.000	3932.500	361.202

Table of compression testing results used for Sigma Plot 9.0 in comparing the 40% Harvested Femurs and the Whole Femurs:

Specimen	Whole/ 40% Harvested	Failure Force (N)
2017	Whole	3460
2000	Whole	2350
1975	Harvested	4182
2031	Harvested	3270
2020	Harvested	5270
2009	Whole	4850
2009	Whole	5250
2028	Harvested	1250

Comparison of the 40% harvested femur versus the whole femur using a one-tailed t-test:

t-test Whole vs 40% harvested

Data source: Data 1 in Notebook 1

Normality Test: Passed (P = 0.585)

Equal Variance Test: Passed (P = 0.781)

Group Name	N	Missing	Mean	Std Dev	SEM
Whole femur	7	3	3977.500	1328.793	664.397
40% harvested	8	4	3493.000	1704.233	852.116

Difference 484.500

t = 0.448 with 6 degrees of freedom. (P = 0.670)

95 percent confidence interval for difference of means: -2159.439 to 3128.439

The difference in the mean values of the two groups is not great enough to reject the possibility that the difference is due to random sampling variability. There is not a statistically significant difference between the input groups (P = 0.670).

Power of performed test with alpha = 0.050: 0.050

The power of the performed test (0.050) is below the desired power of 0.800. Less than desired power indicates you are more likely to not detect a difference when one actually exists. Be cautious in over-interpreting the lack of difference found here.

Appendix I

Image Analysis for Stress-Strain

Take TIFF format images of samples for analysis

Open Scion Image 1.63 for MacOS 7.5 to 9.x. Acquisition and Analysis Software (Scion Corporation, Maryland).

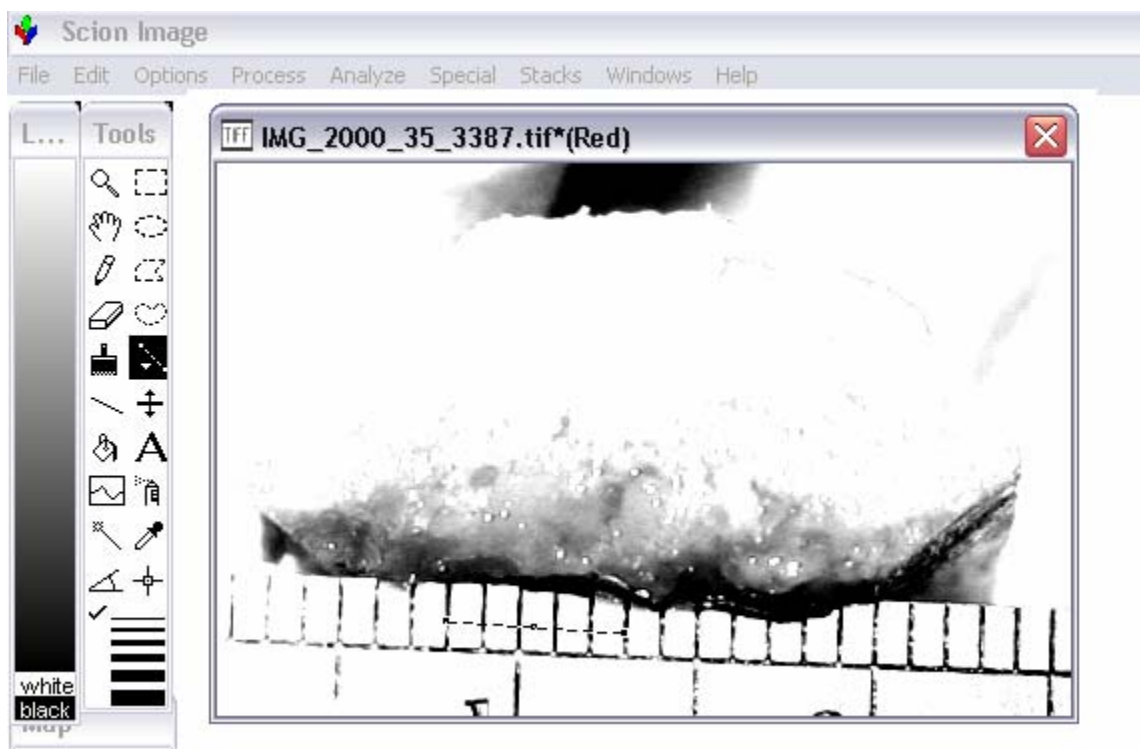
File >> Import.

Import the image that needs to be analyzed.

Calibration:

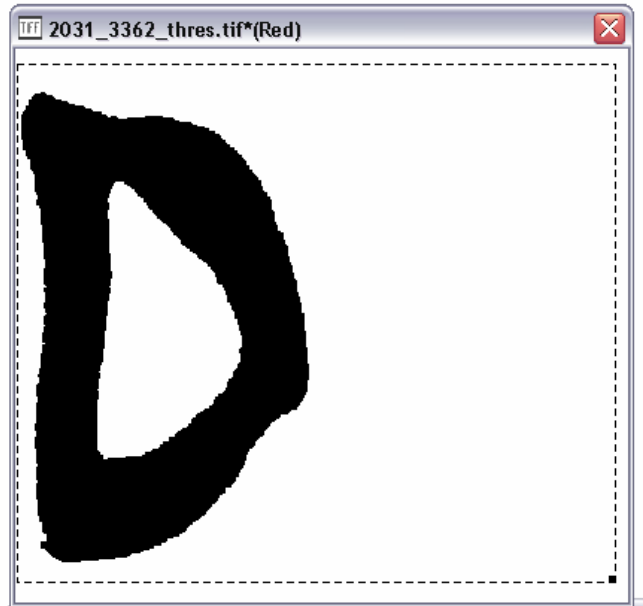
Images should have a reference to make proper dimensional calibrations for analysis. Use the dashed line (highlighted below) to measure the reference in the image.

Analyze >> Calibrate Type in the length that the reference is measuring (e.g. the below image is referenced by a 5mm measurement). This creates a pixels per mm ration.



To measure the area of cortical bone, threshold the picture so that the background is white and the cortical bone is black.

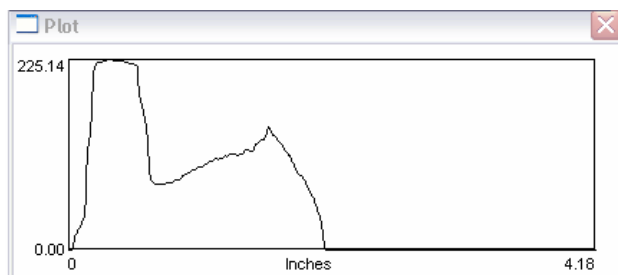
Options >> Threshold



Area Profiles:

To analyze the area profile of the sample, encompass the image with a dashed rectangle (refer to image above). To make sure that the appropriate axis of the image is set as the x-axis of the profile plot, make the x-axis longer than the y-axis as shown in figure above. Once the rectangle is in place, follow these instructions.

Analyze >> Plot Profile (the below plot will appear)



Exportation of Data:

Keep the plot highlighted and follow the next directions.

File >> Export Export the data set to a folder that can be easily found and remembered.

Tecplot version 10.0 release 3 software (Tecplot Incorporated, Washington)

Importing Data:

File >> Load Data File(s) Find the data set that you saved earlier

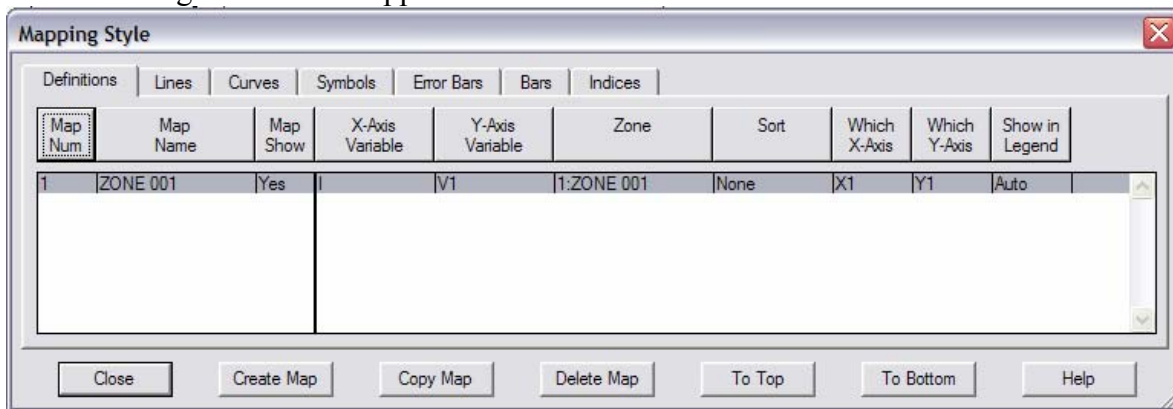
The following window will appear, just click the OK button if the Initial Plot Type is XY Line.



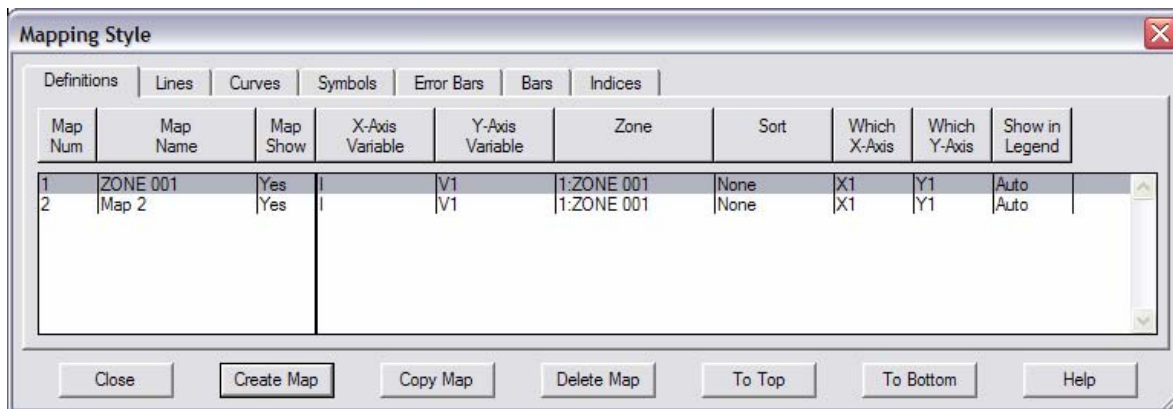
A graph of the data set will appear.

Curve Fitting:

Click on the “Mapping Style...” button in a toolbar on the left side of the screen. The following window will appear:

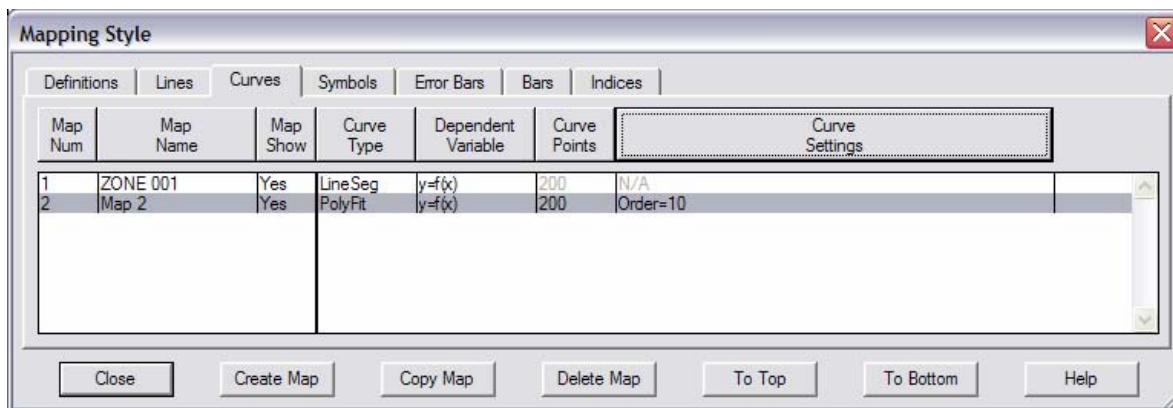


Create Map >> OK (in the window that appears)

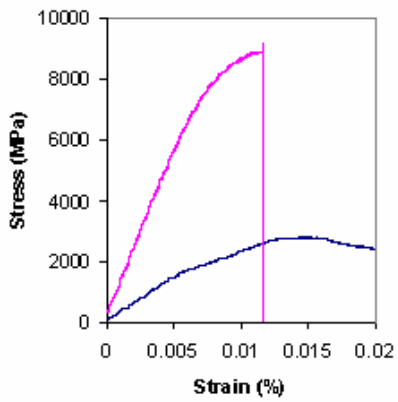


Curves >> Curve Type (make sure to have Map 2 highlighted) >> Polynomial Fit

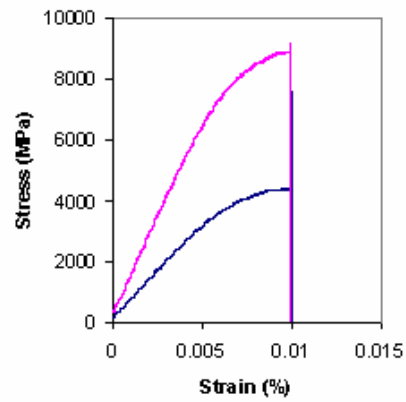
Curve Settings >> Change the order of the curve from 3 to 10.



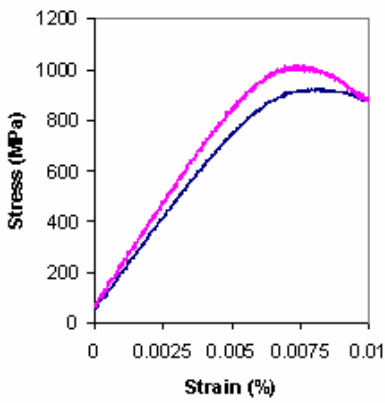
Stress-Strain Results



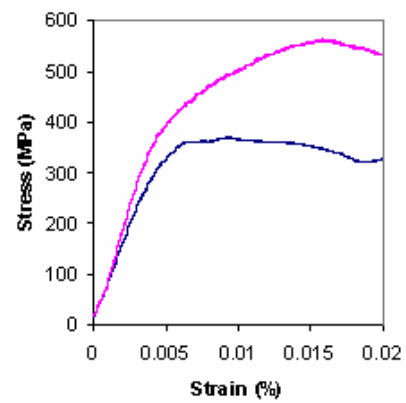
— 2000 Average
— Standard Deviation



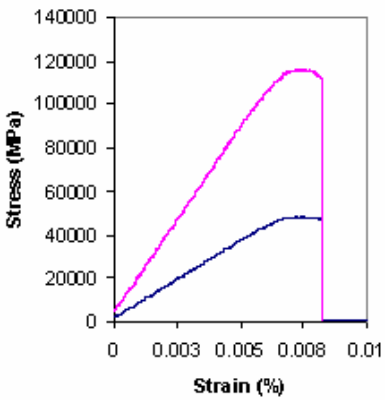
— 2008 Average
— Standard Deviation



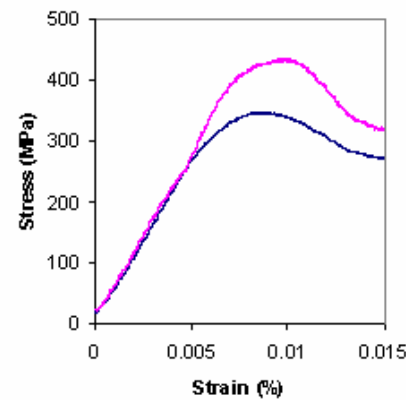
— 1975 Average
— Standard Deviation



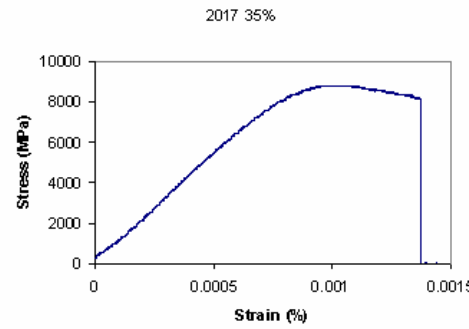
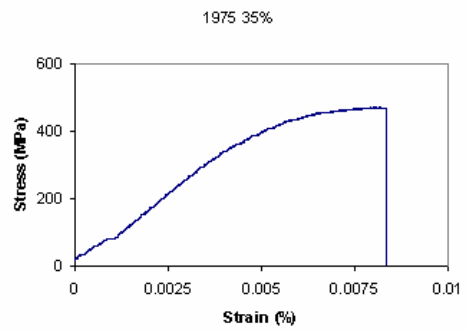
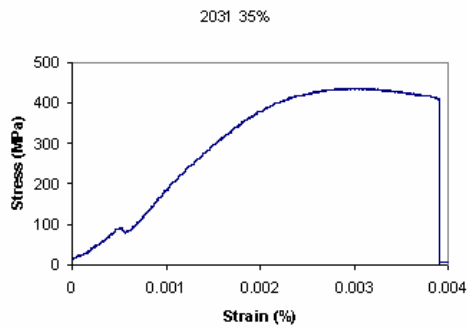
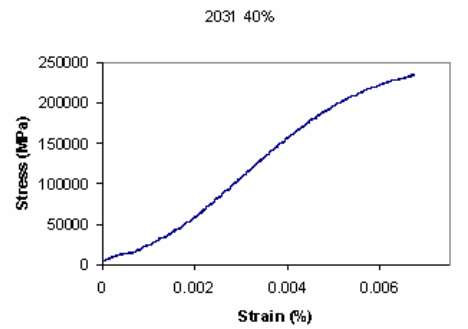
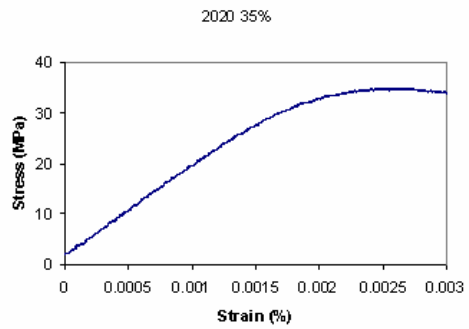
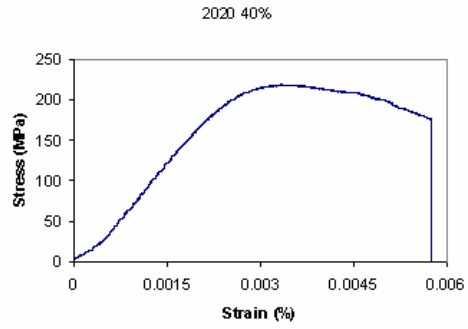
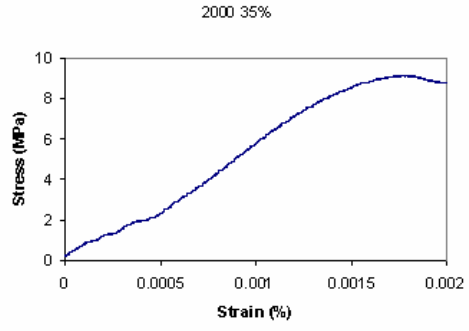
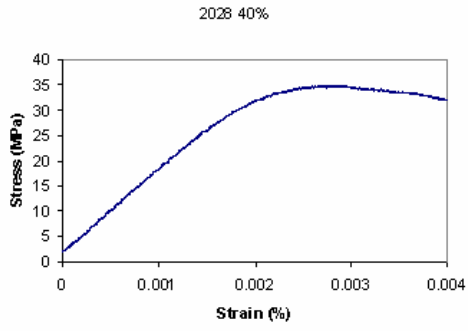
— 2031 Average
— Standard Deviation

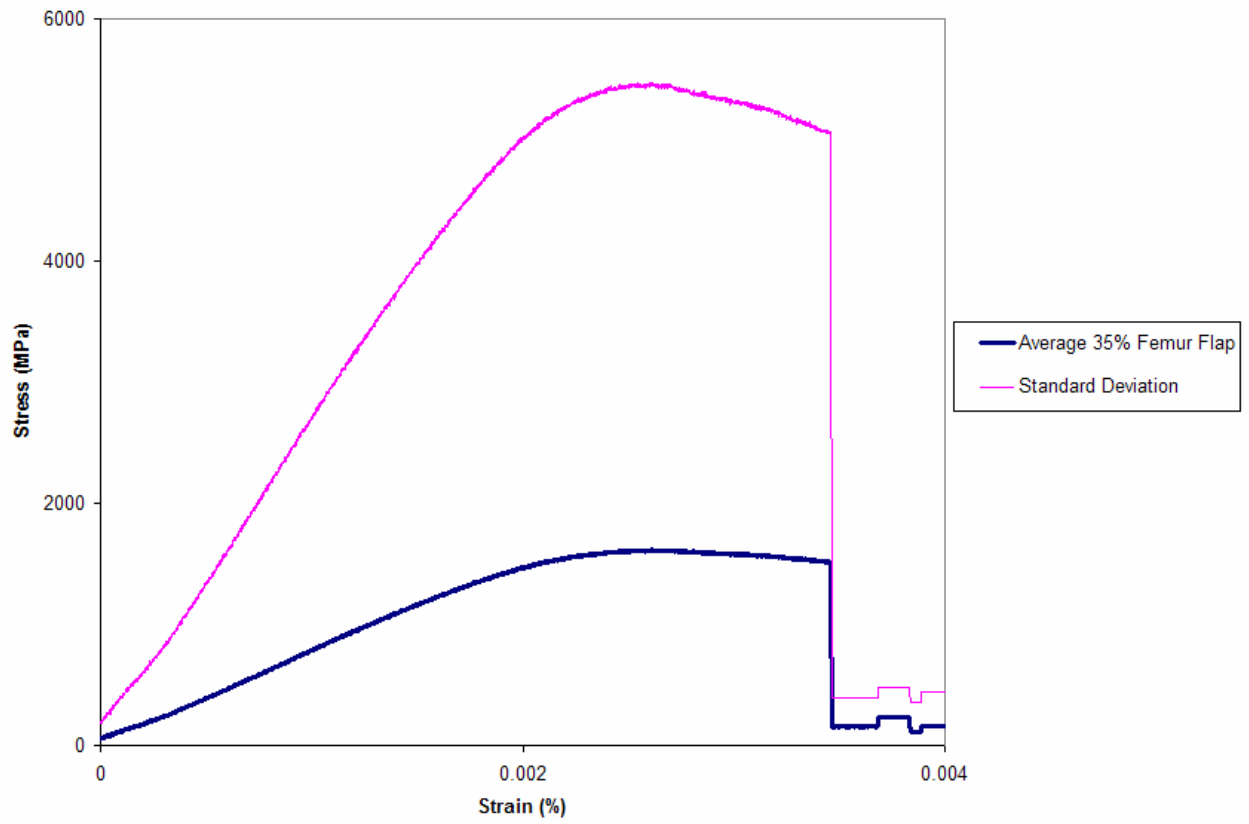


— 2017 Average
— Standard Deviation



— 2028 Average
— Standard Deviation

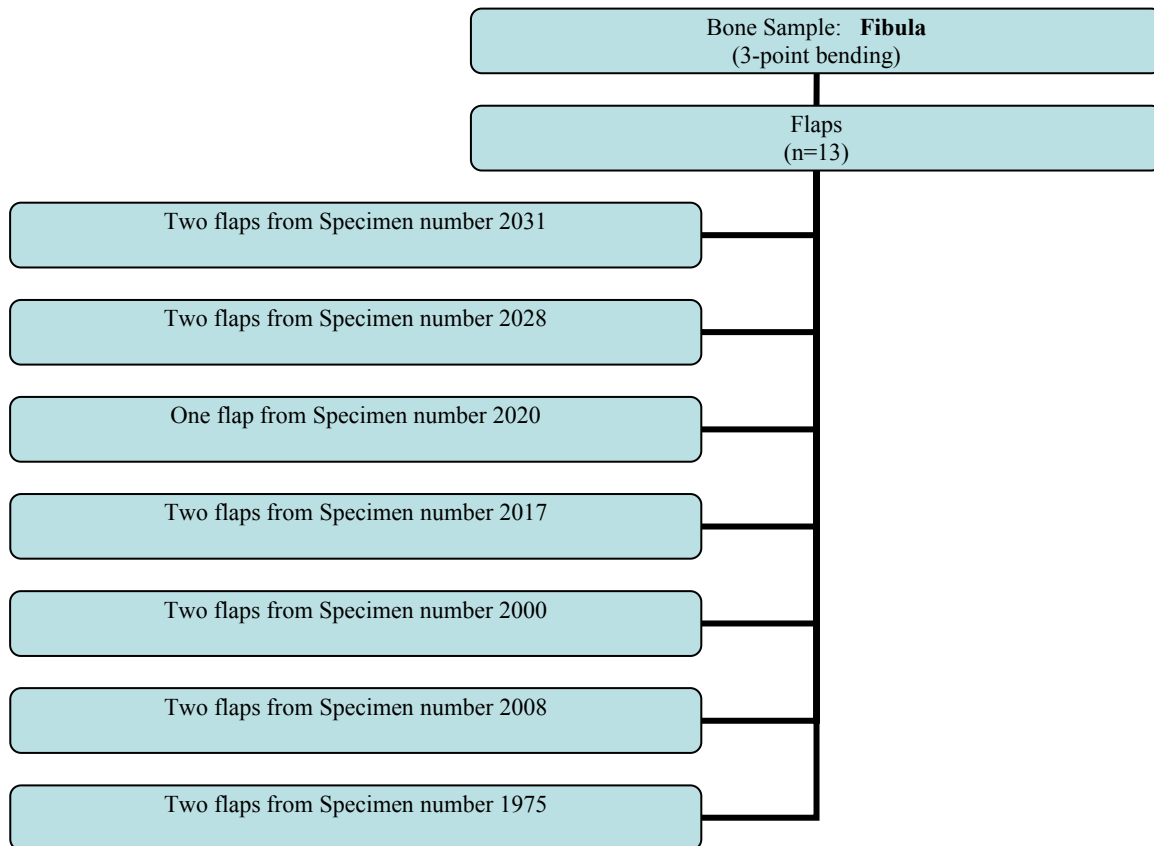




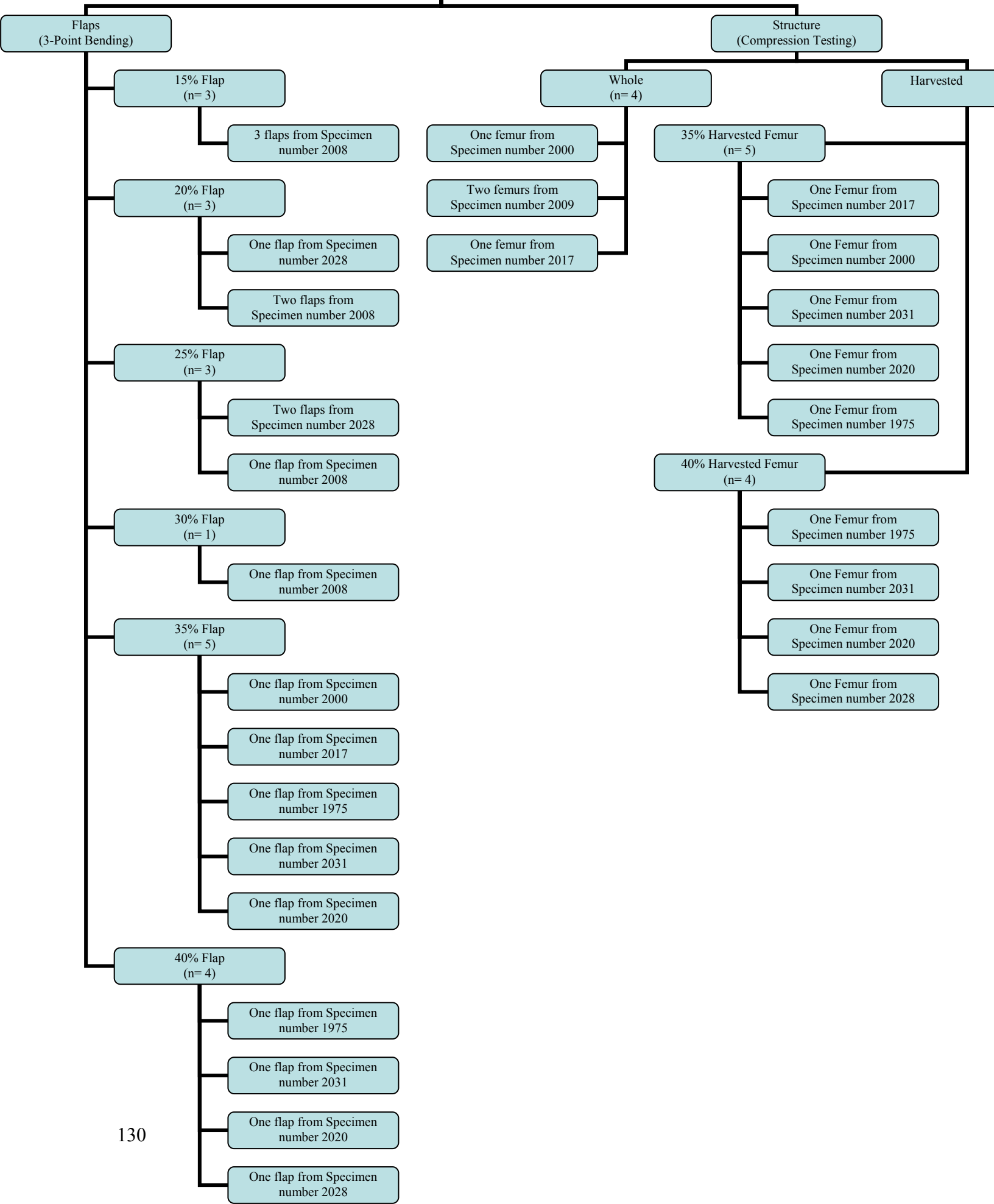
Appendix J

How the bones were broken up for testing:

The following are diagrams of which cadavers were used in the 3-point bending and compression tests conducted on the fibulas and the femurs, respectively.



Bone Sample:
Femur



Appendix K

Individual's Tags:

Table of Descriptions of Individual Cadavers Used in Testing:

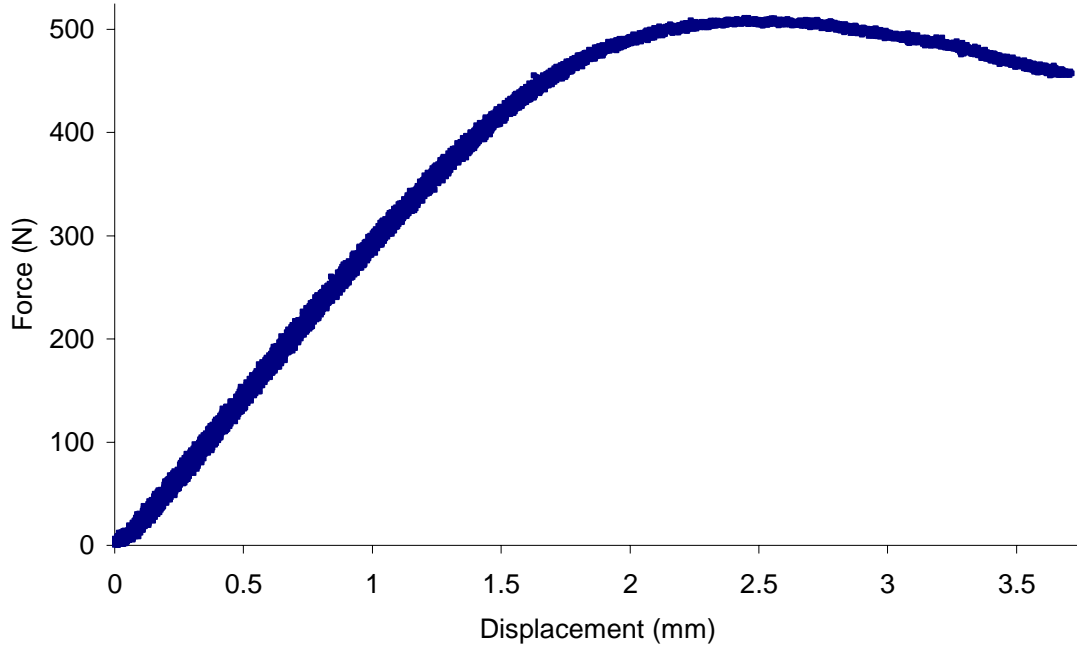
Individual's Unit Number	Sex	Age	Cause of Death
1975	F	77	Chronic Obstructive Lung Disease
2000	M	86	Dementia/Parkinson's Disease
2008	F	92	C.V.A.
2009	M	85	Respiratory Failure
2017	F	95	C.V.A.
2020	M	94	Cardiac Arrest
2028	F	89	Dehydration/Alzheimer's
2031	M	85	Pneumonia

This is a list of all the individuals that provided bone samples in order to complete this MQP

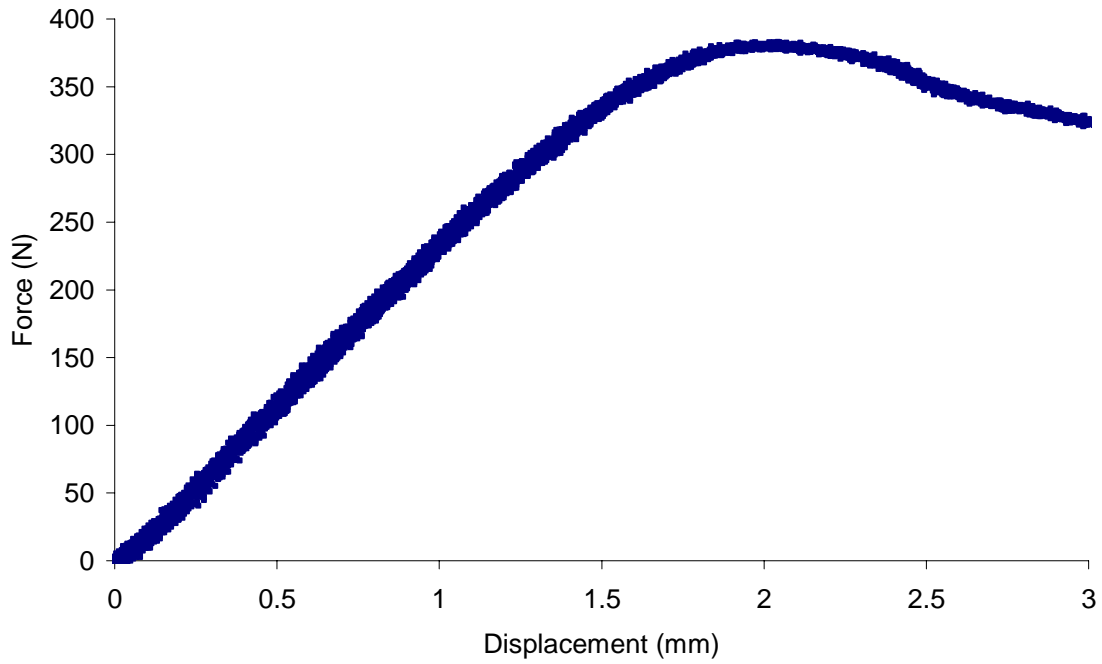
Appendix L

Fibula Three Point Testing

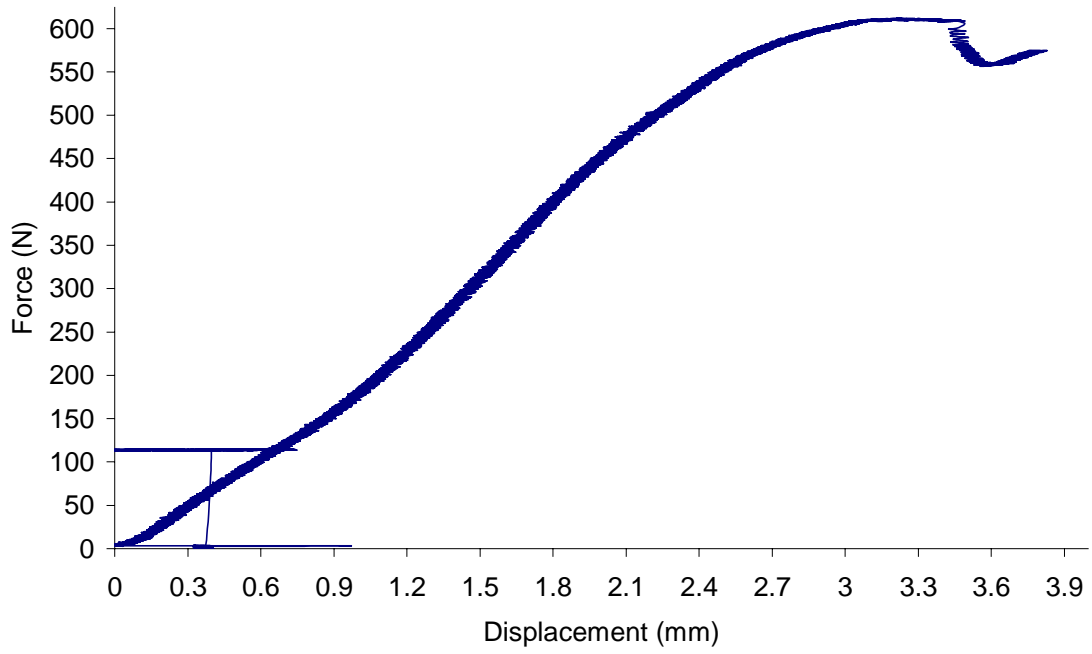
Fibula 1975a



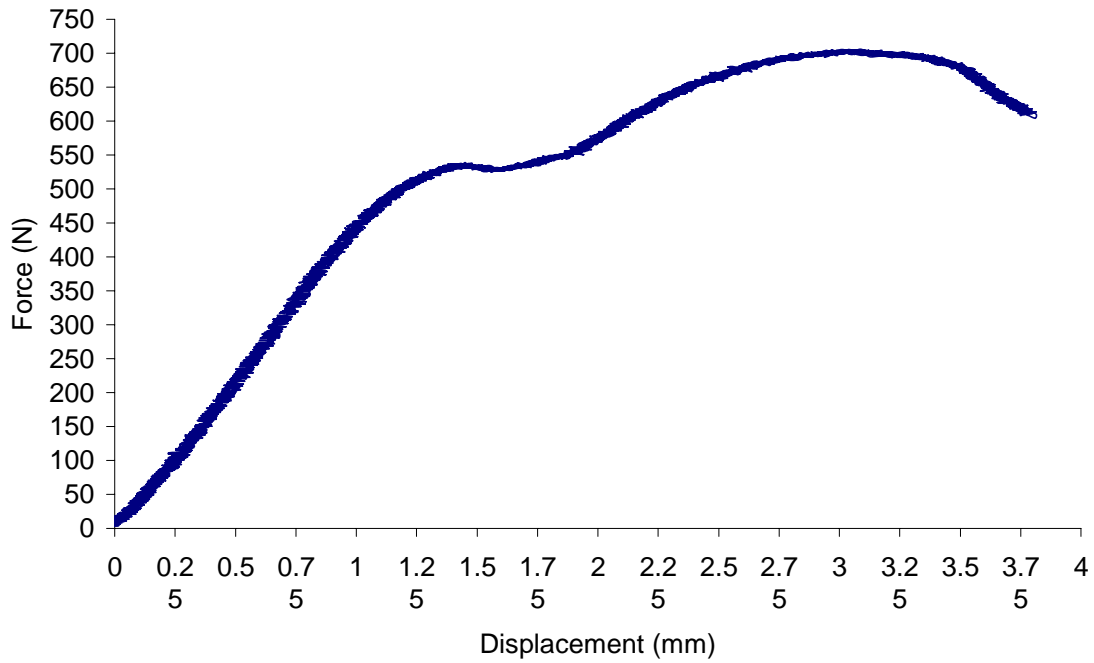
Fibula 1975b



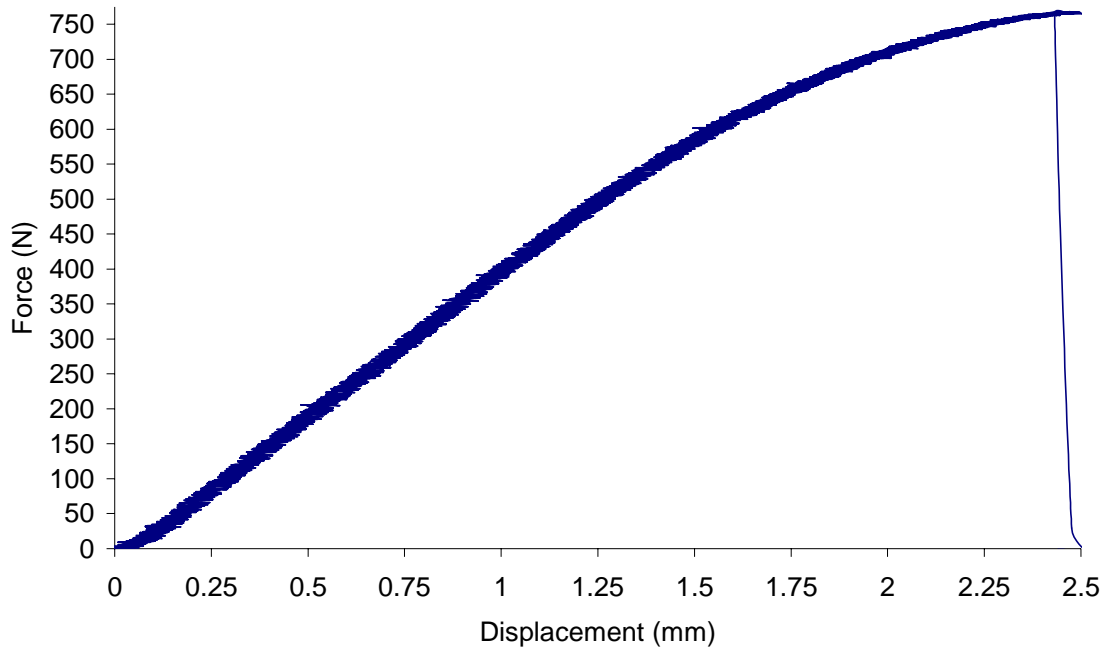
Fibula 2000a



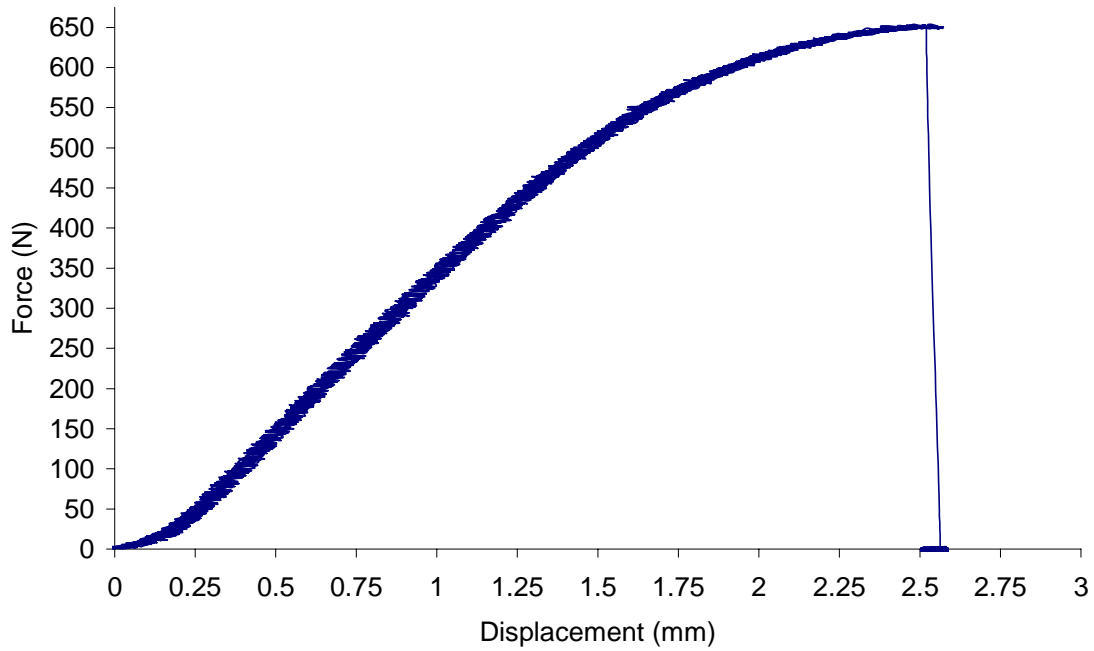
Fibula 2000b



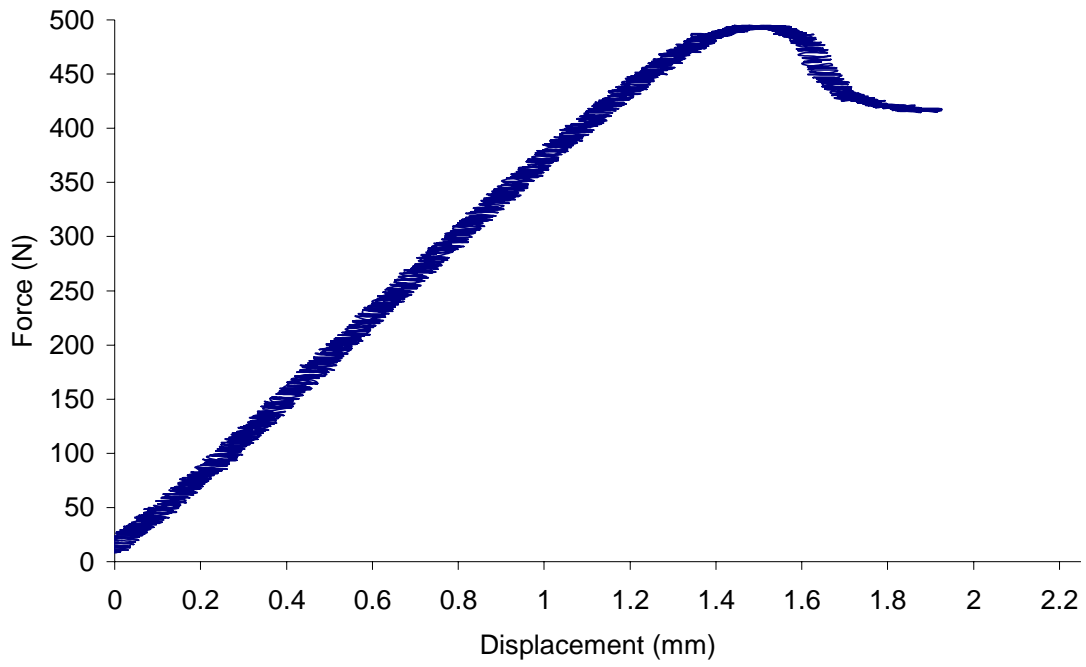
Fibula 2008a



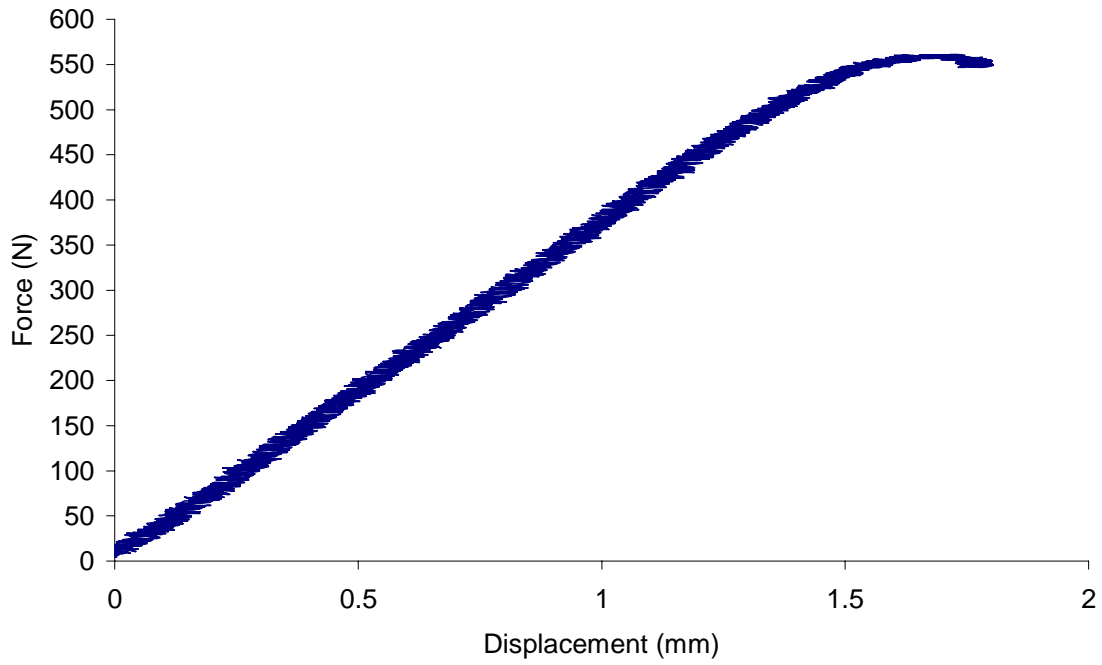
Fibula 2008b



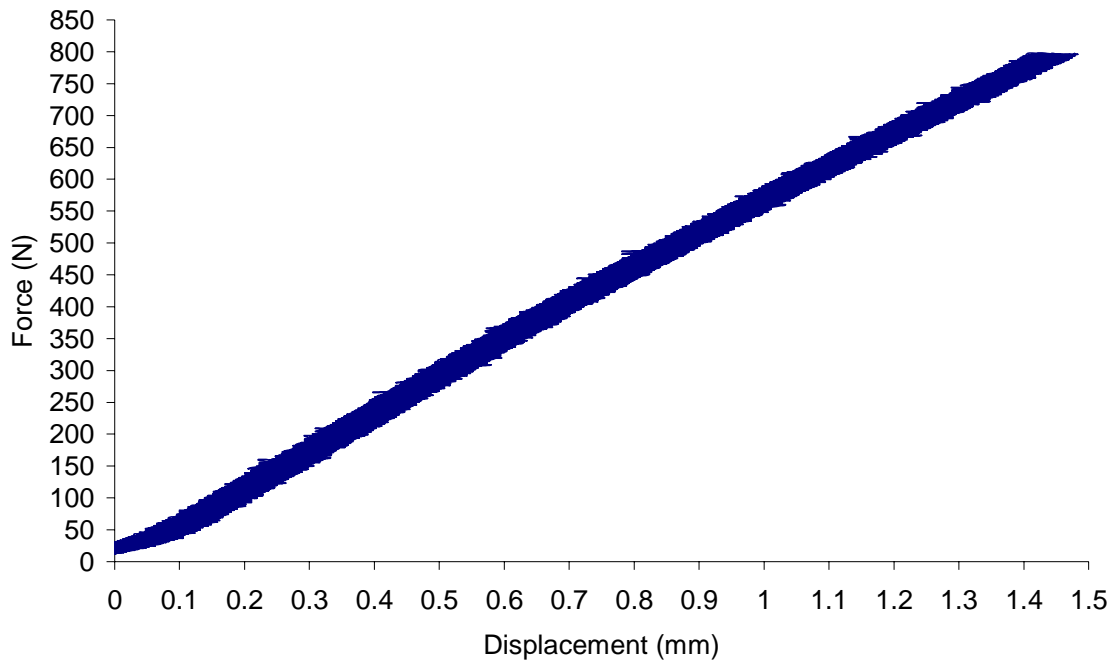
Fibula 2017b



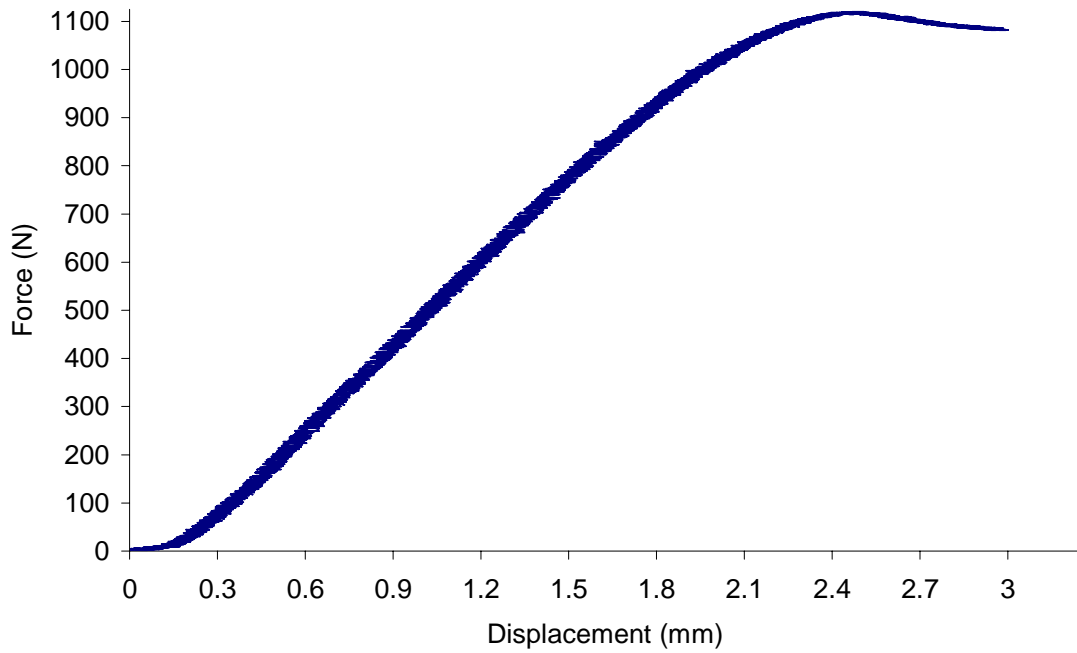
Fibula 2017a



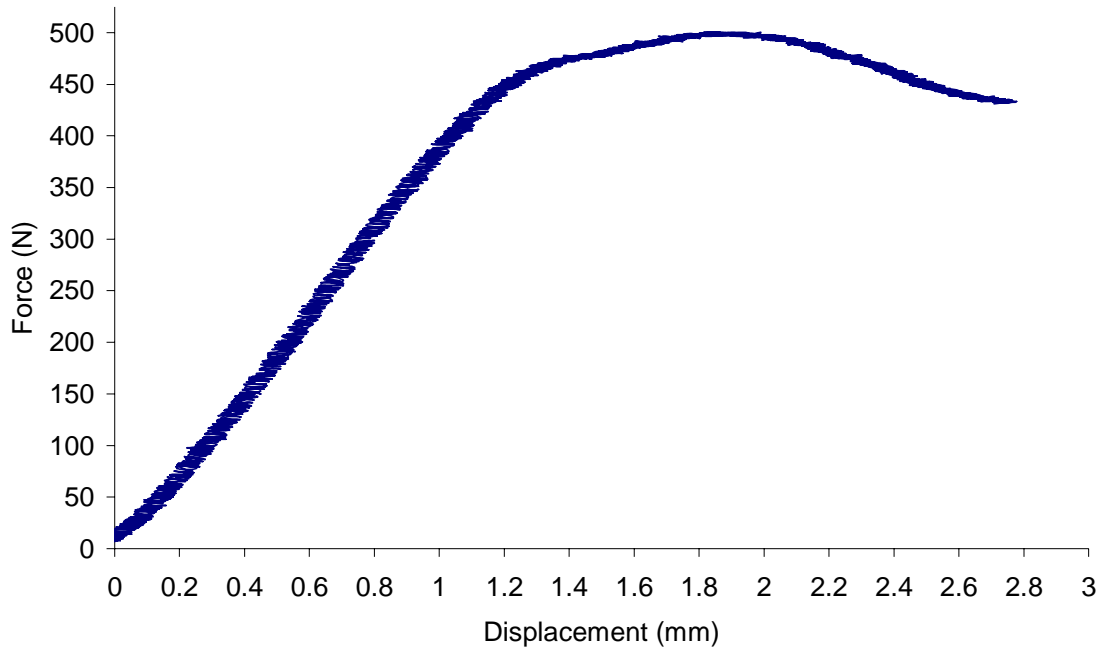
Fibula 2020a



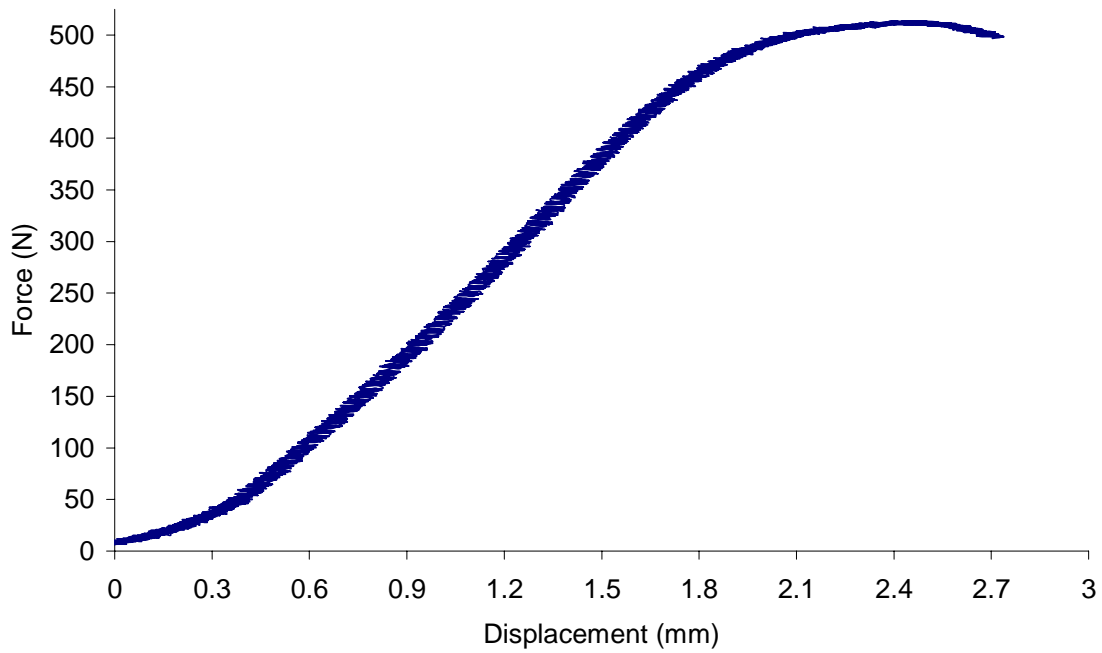
Fibula 2020b



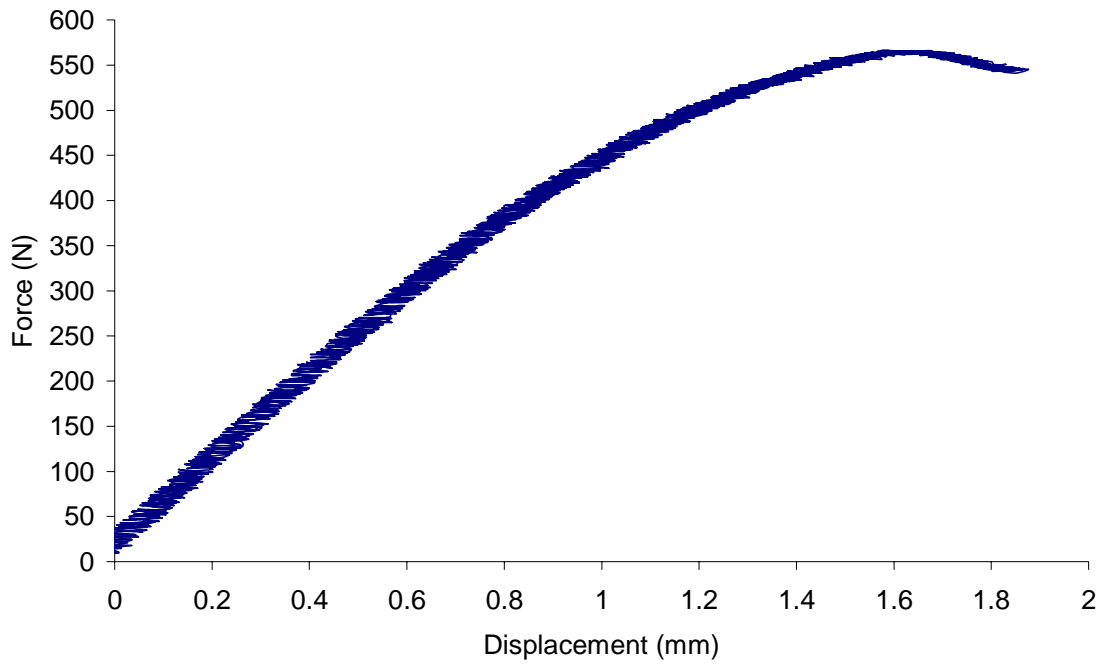
Fibula 2028a



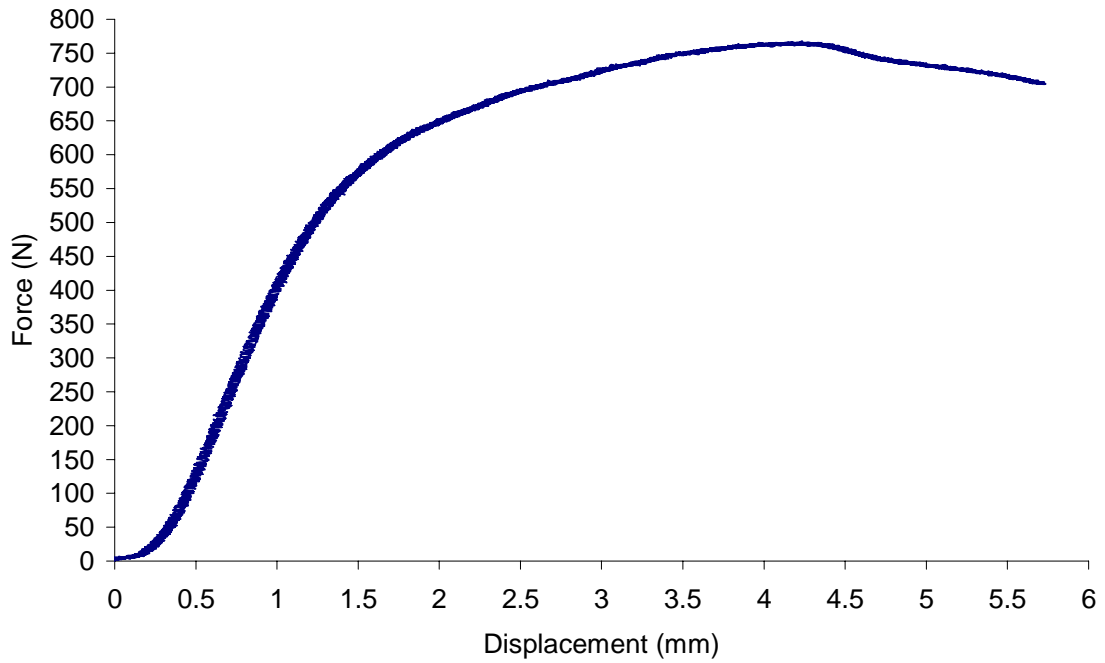
Fibula 2028b



Fibula 2031a

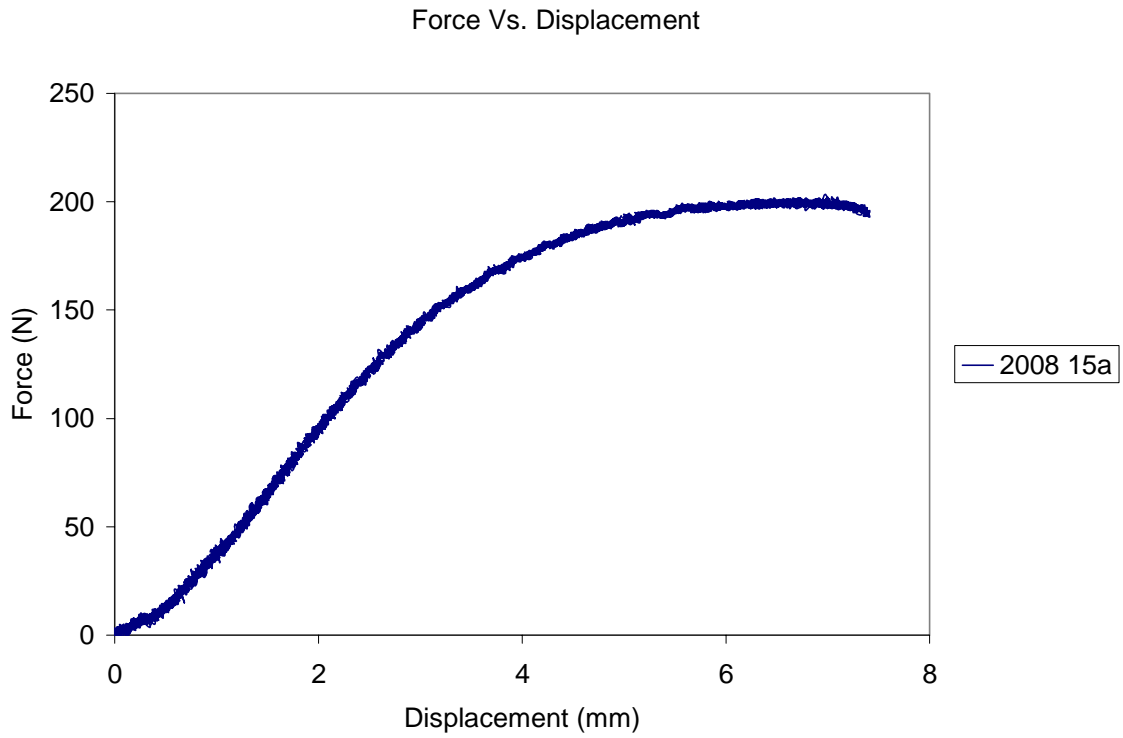


Fibula 2031b

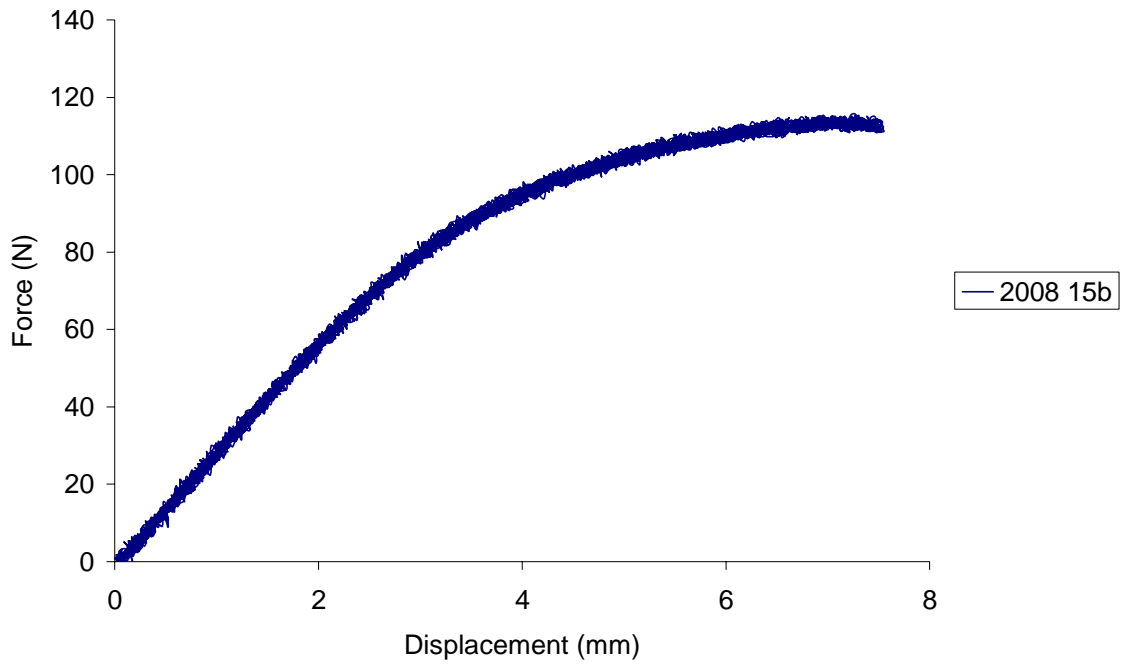


Appendix M

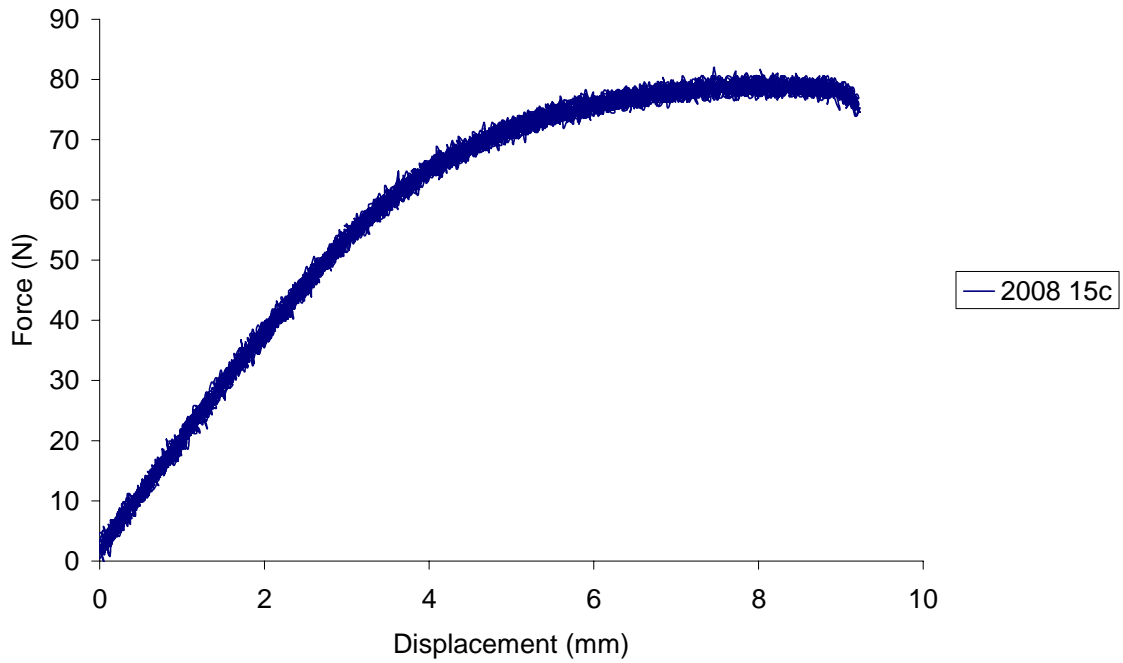
Initial Regional Studies of the Femur Harvest Region



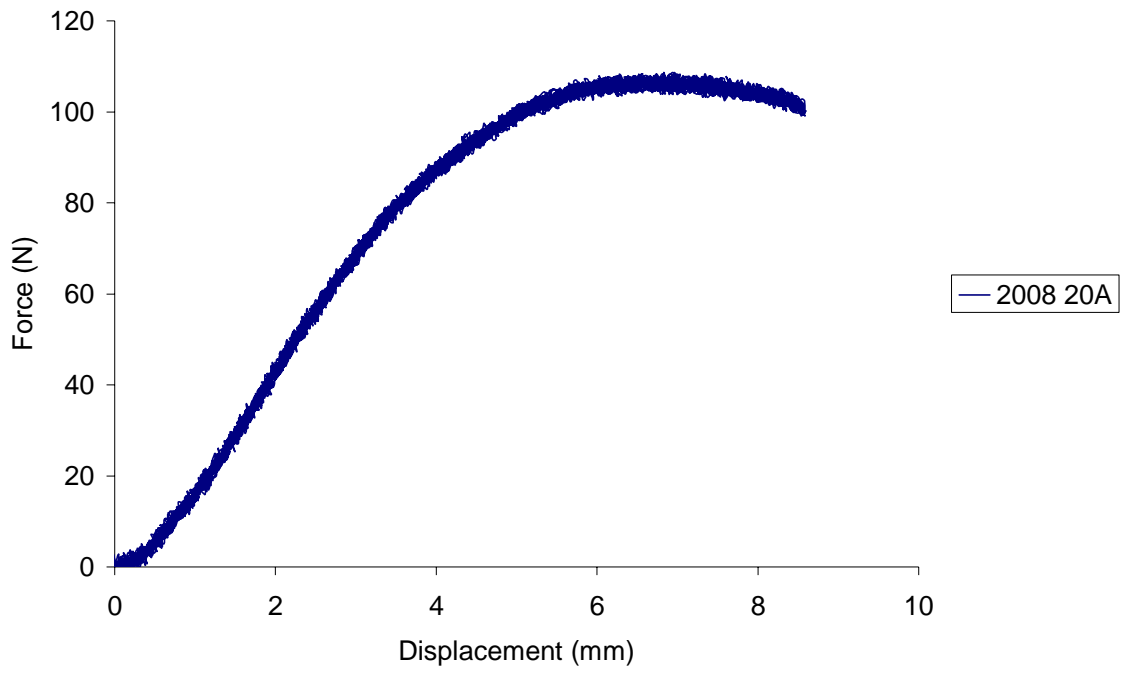
Force Vs. Displacement



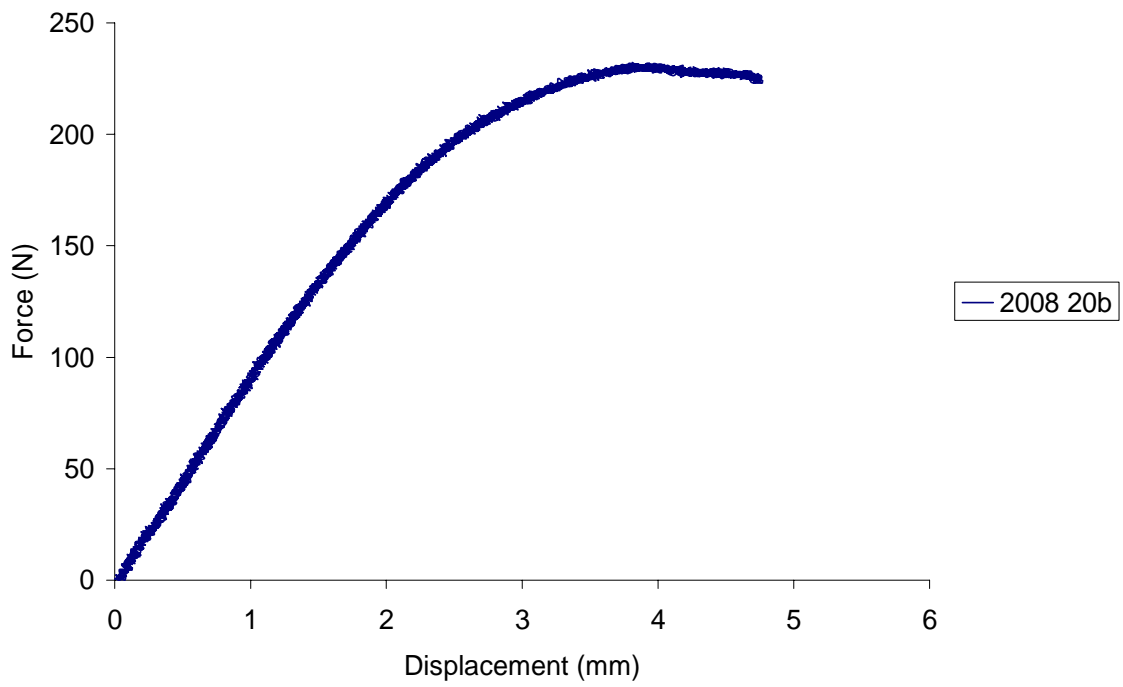
Force Vs. Displacement



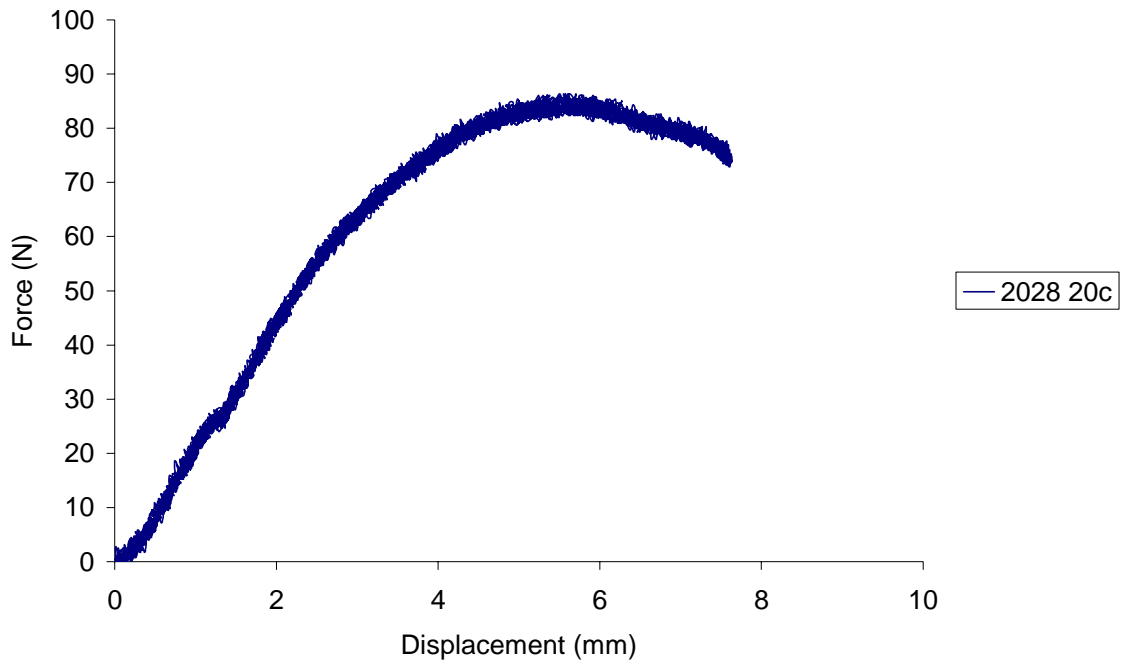
Force Vs. Displacement



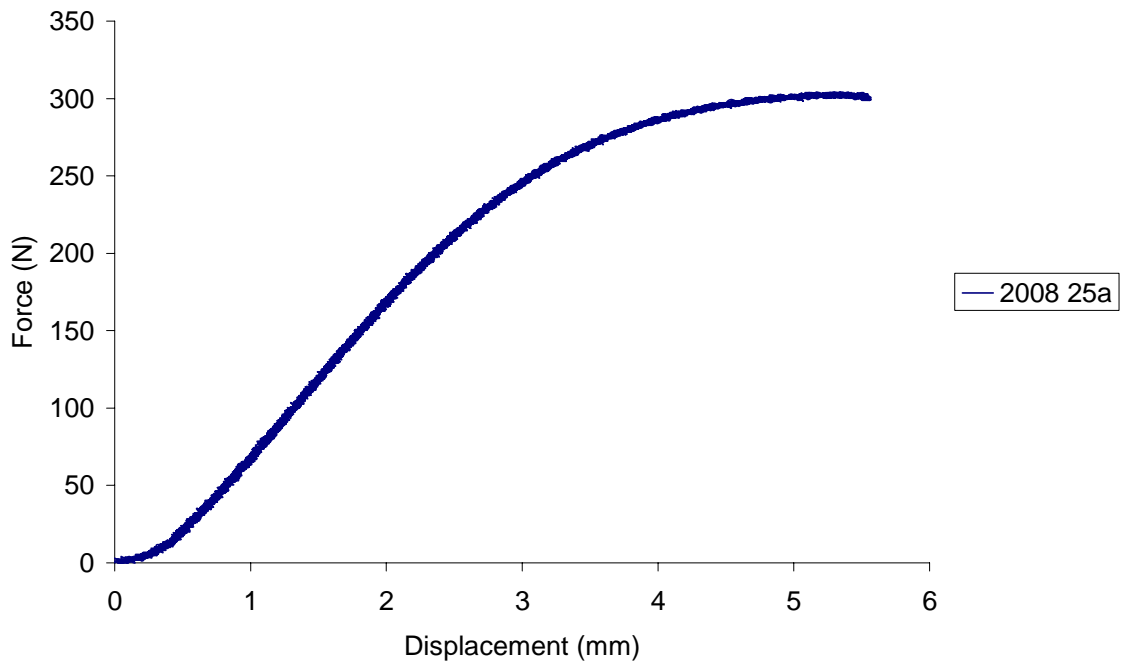
Force Vs. Displacement



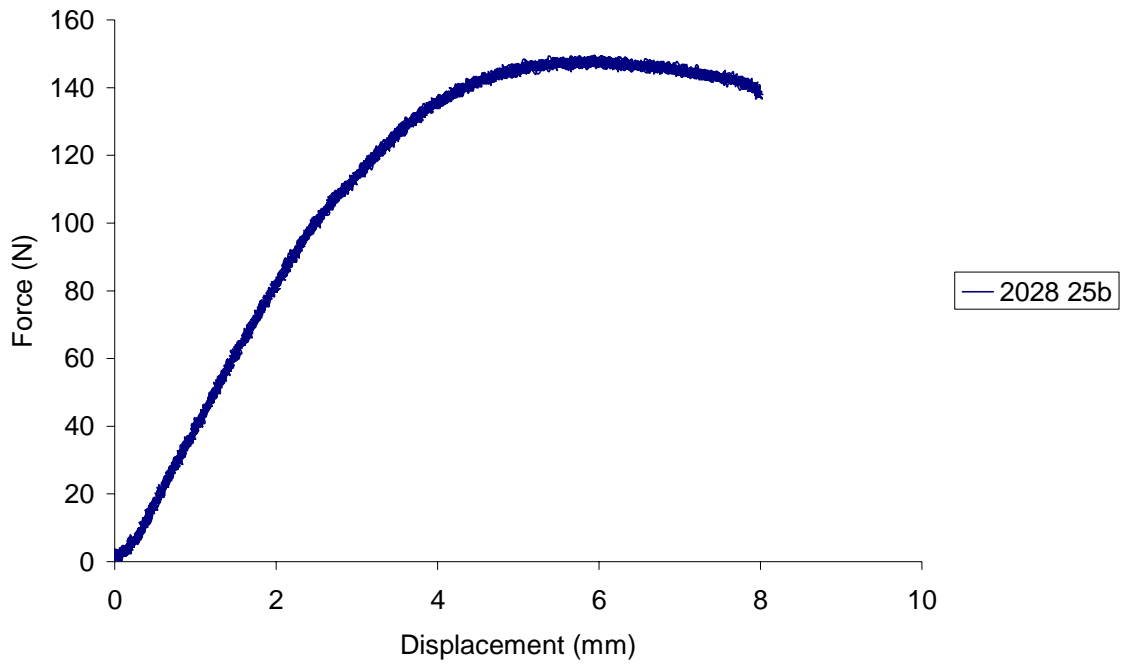
Force Vs. Displacement



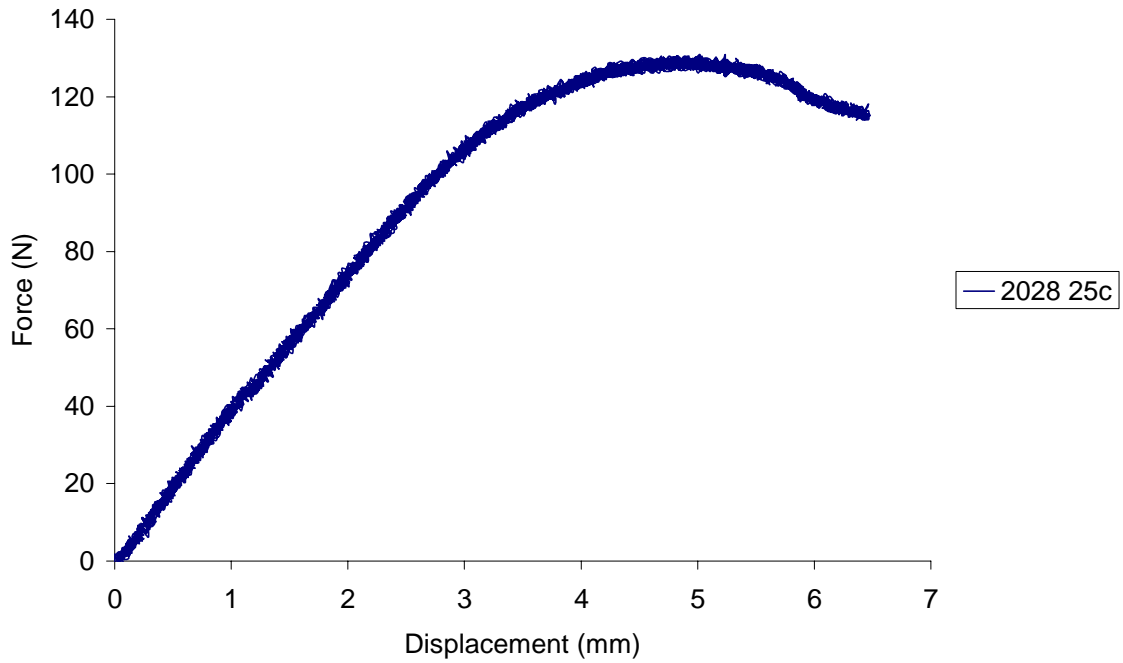
Force Vs. Displacement



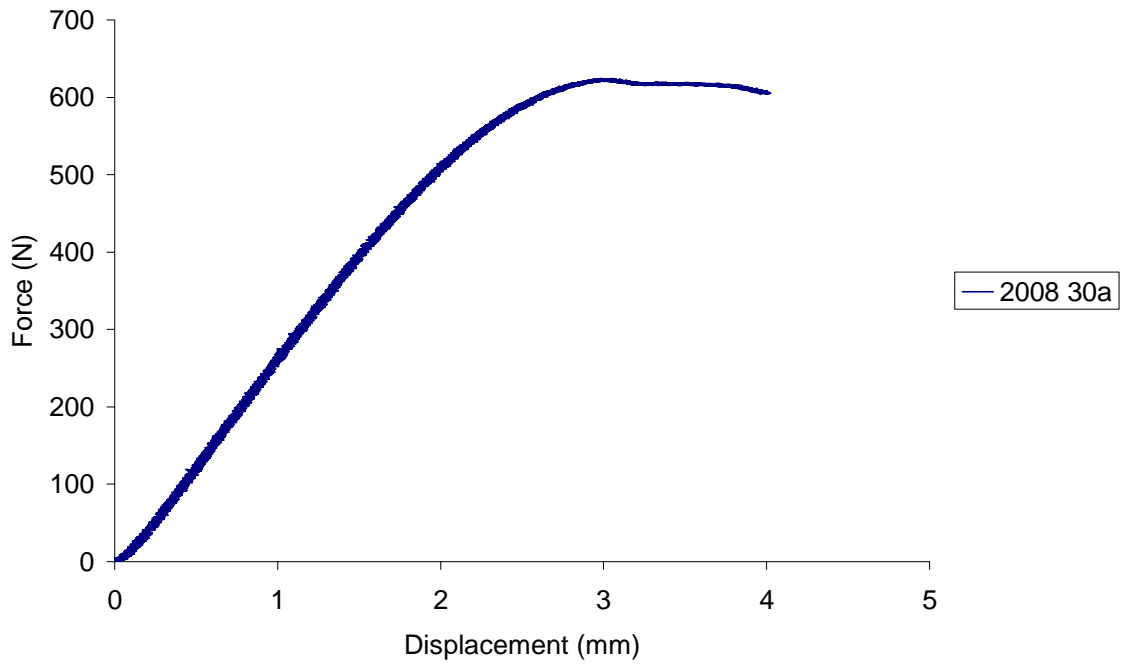
Force Vs. Displacement



Force Vs. Displacement

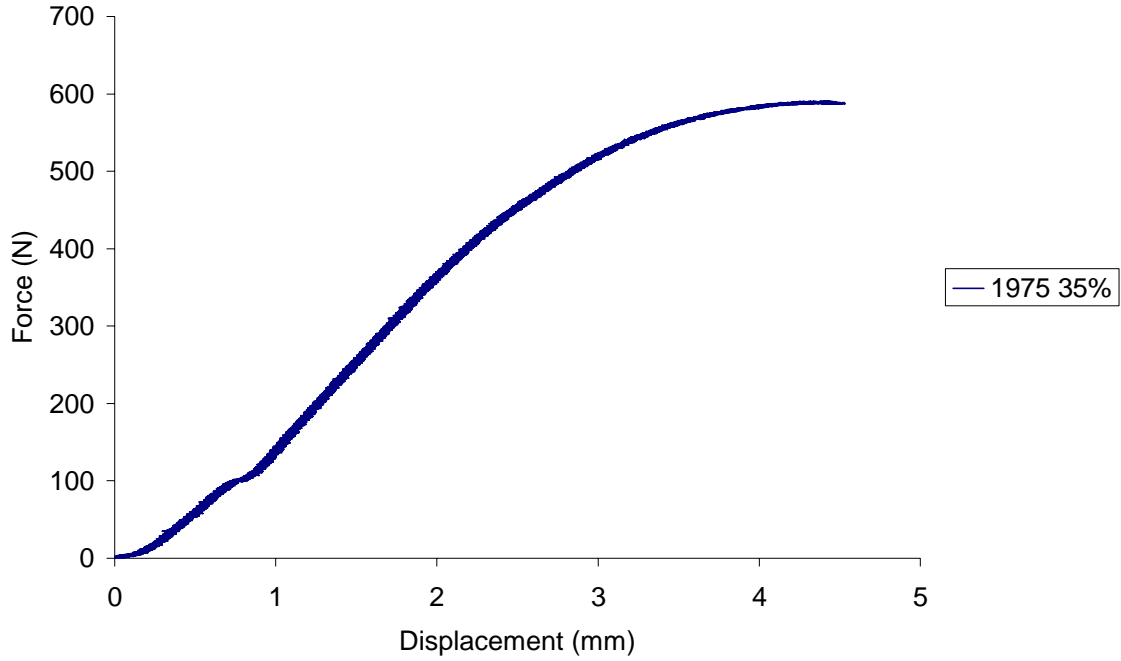


Force Vs. Displacement

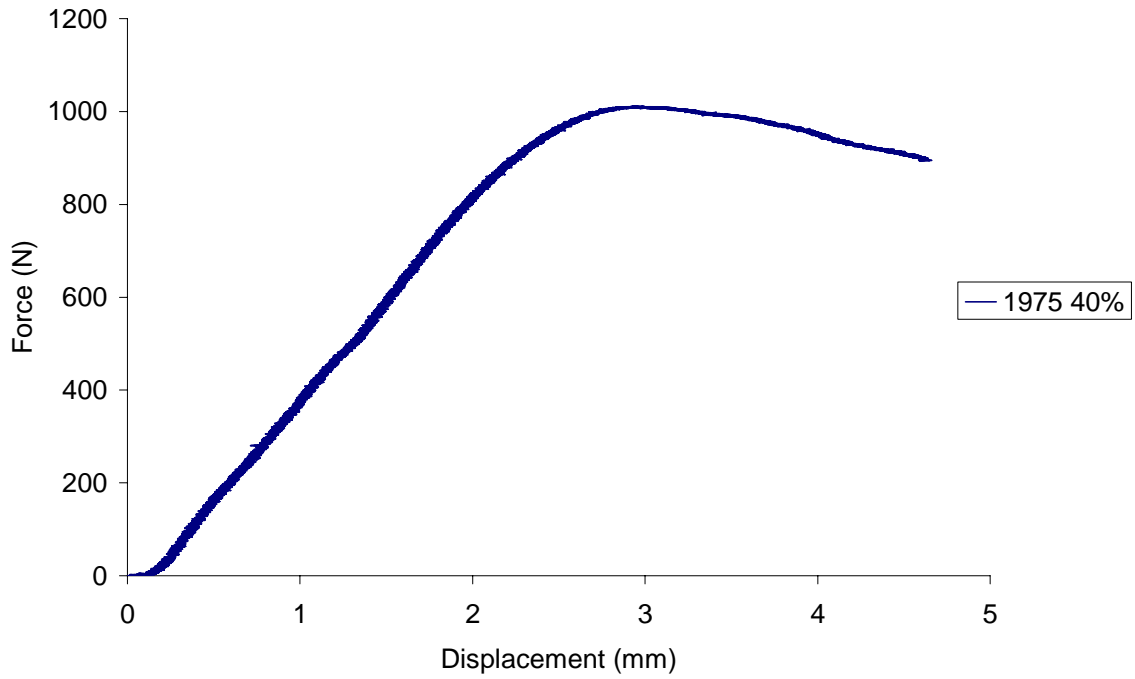


Studies of the 35% and 40% Circumferential Femur Flaps

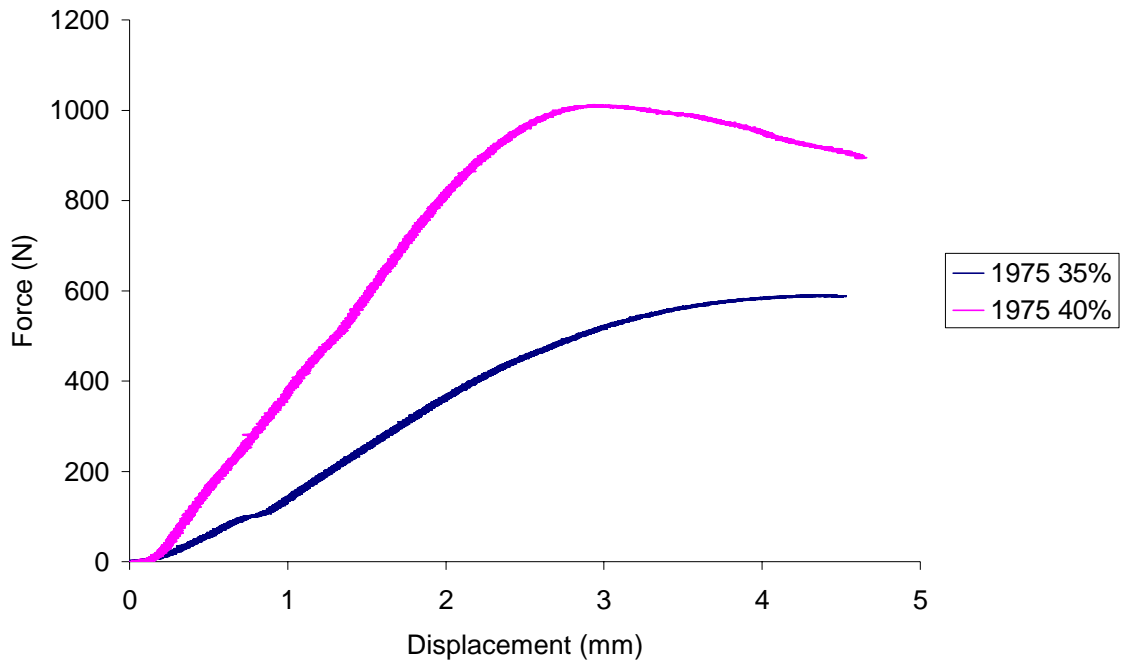
Force Vs. Displacement



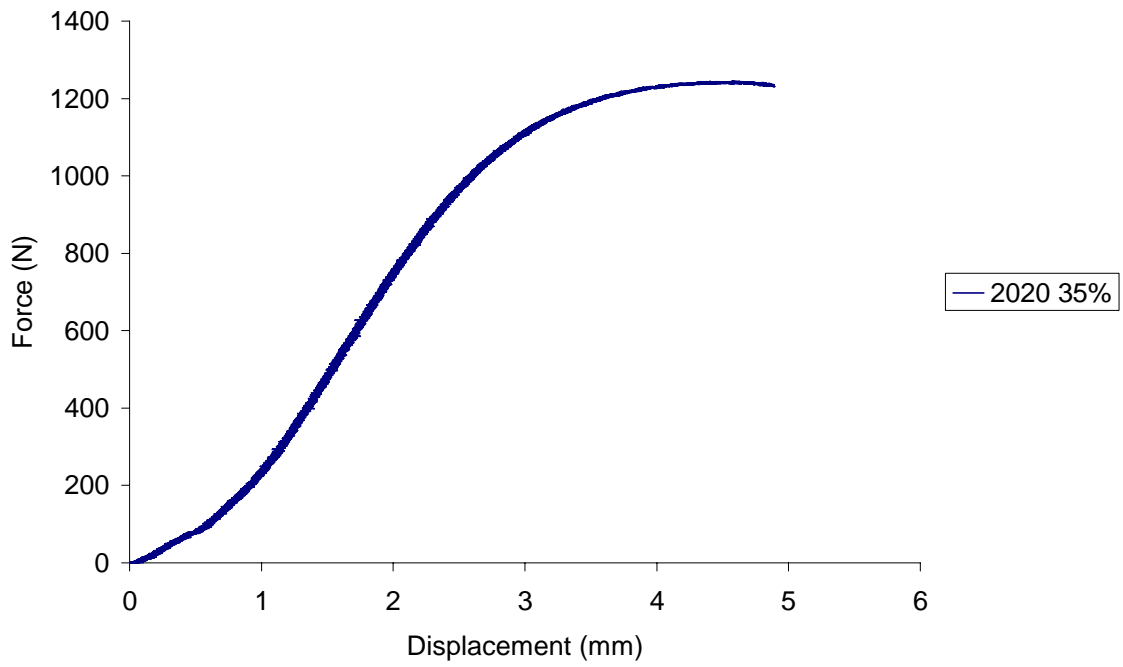
Force Vs. Displacement



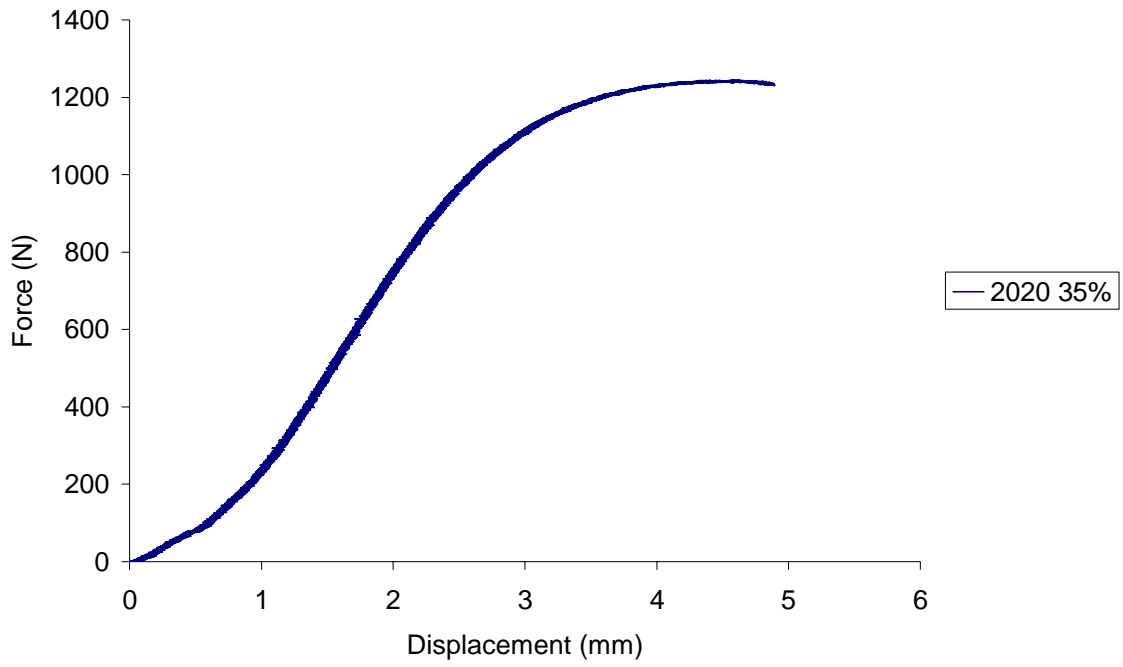
1975 35% Vs 40%



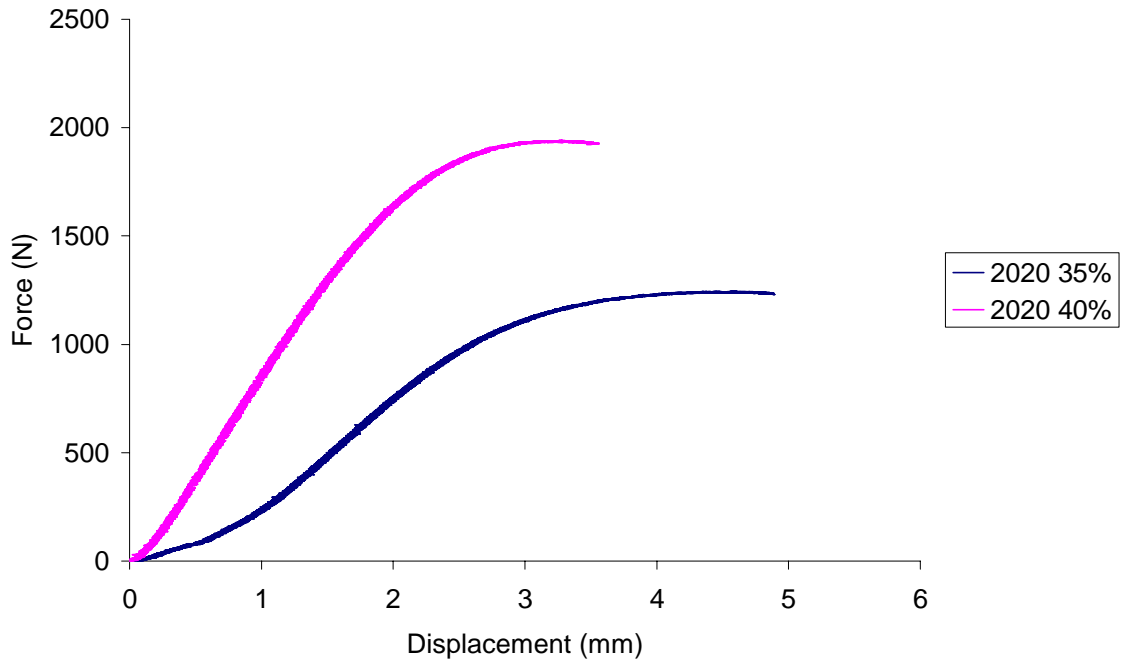
Force Vs. Displacement



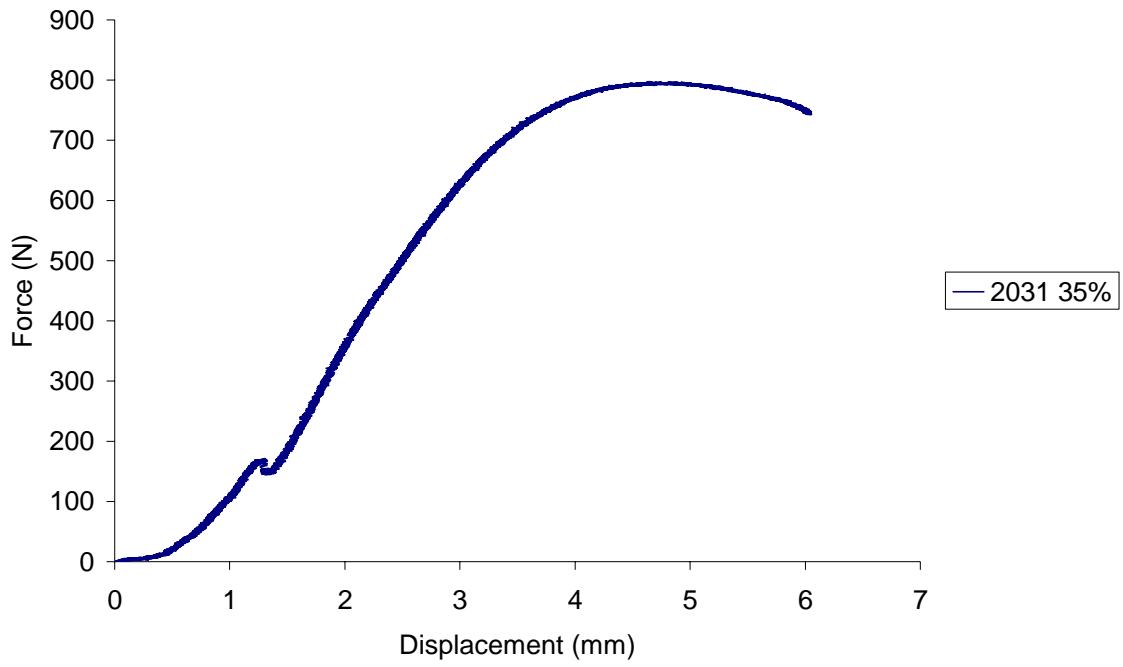
Force Vs. Displacement



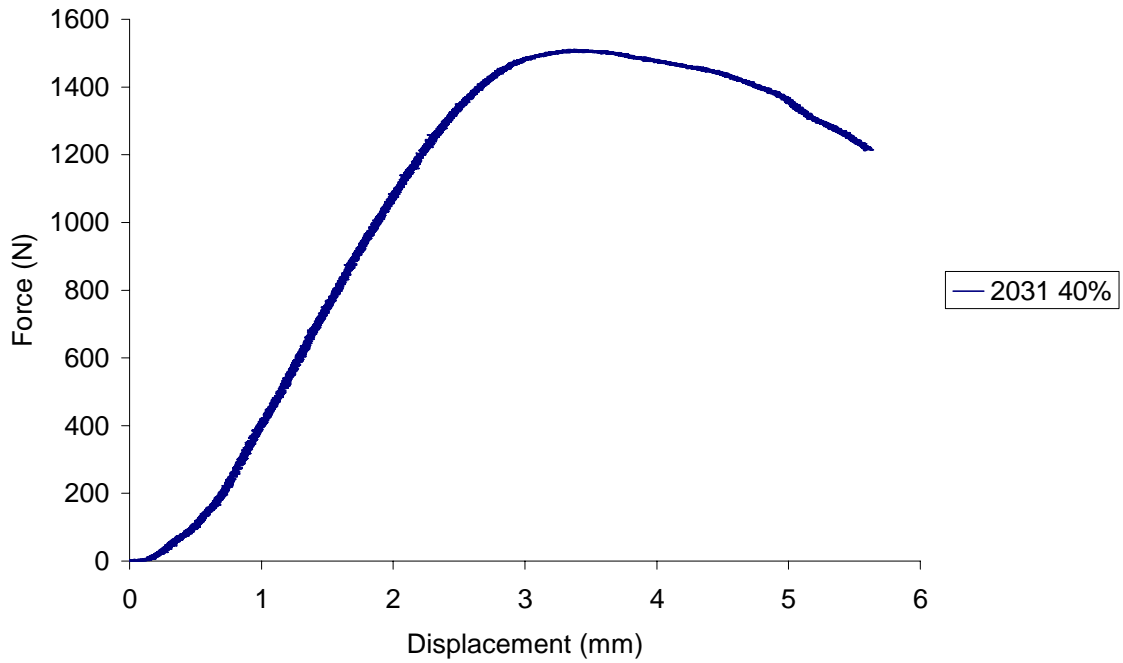
2020 35% Vs. 40%



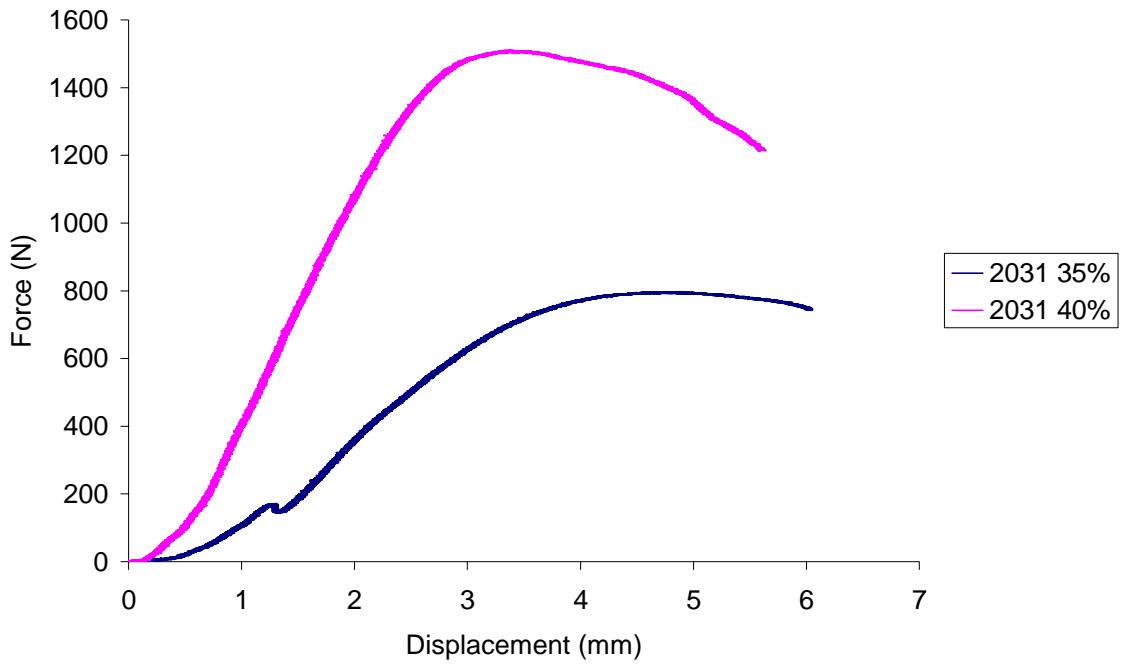
Force Vs. Displacement



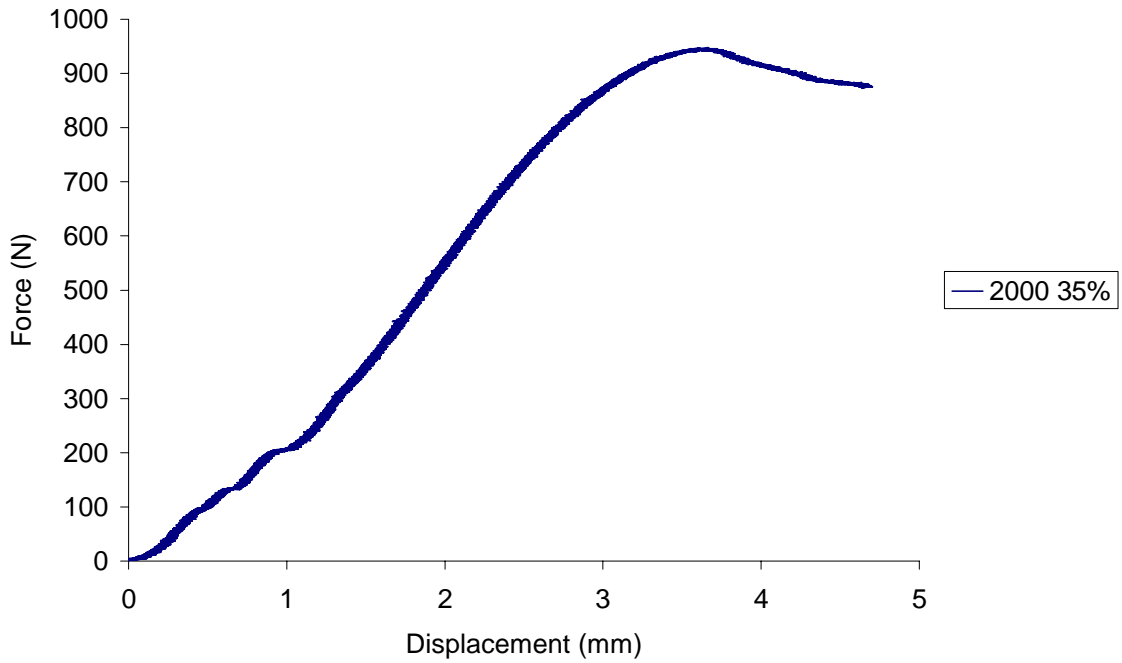
Force Vs. Displacement



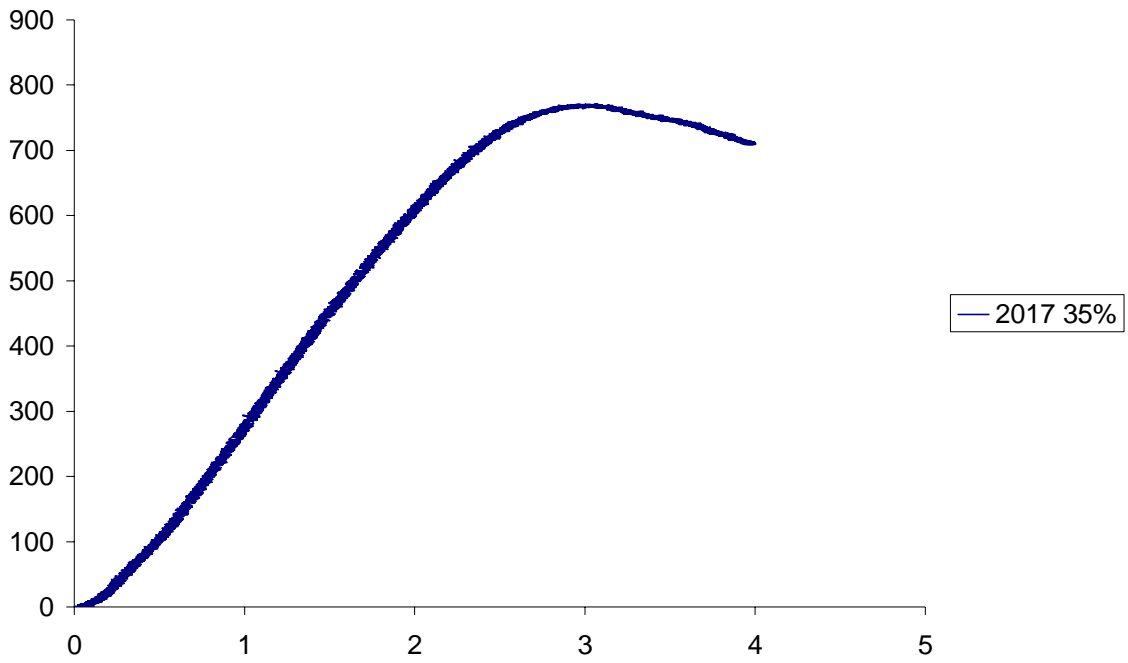
2031 35% Vs. 40%



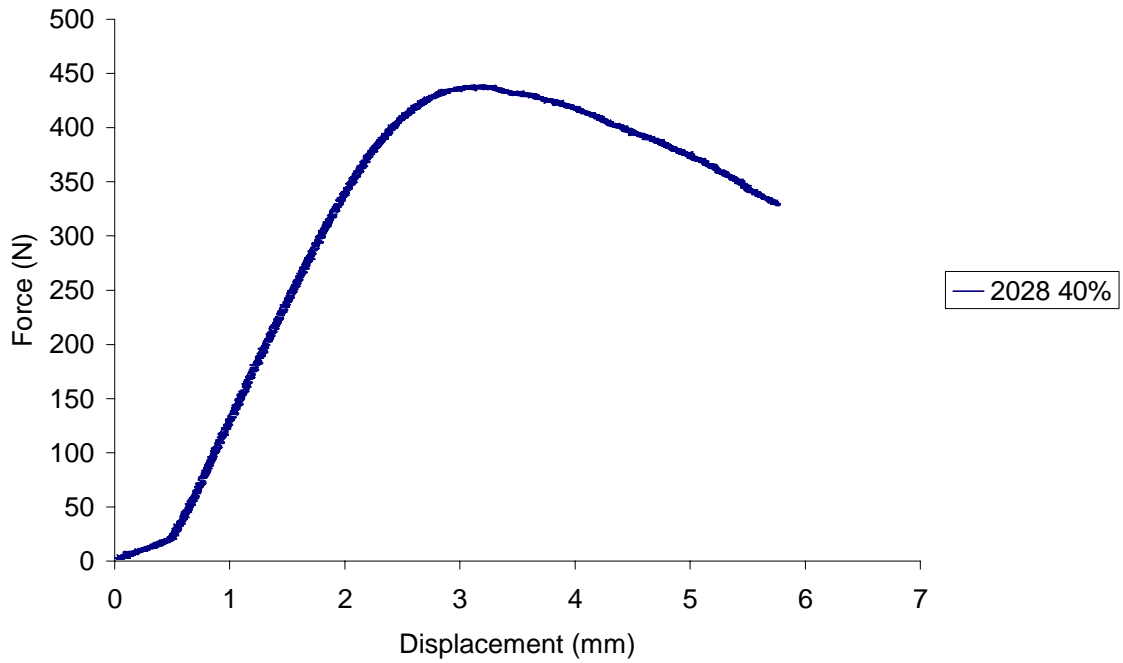
Force Vs. Displacement



Force Vs. Displacement



Force Vs. Displacement



Appendix N

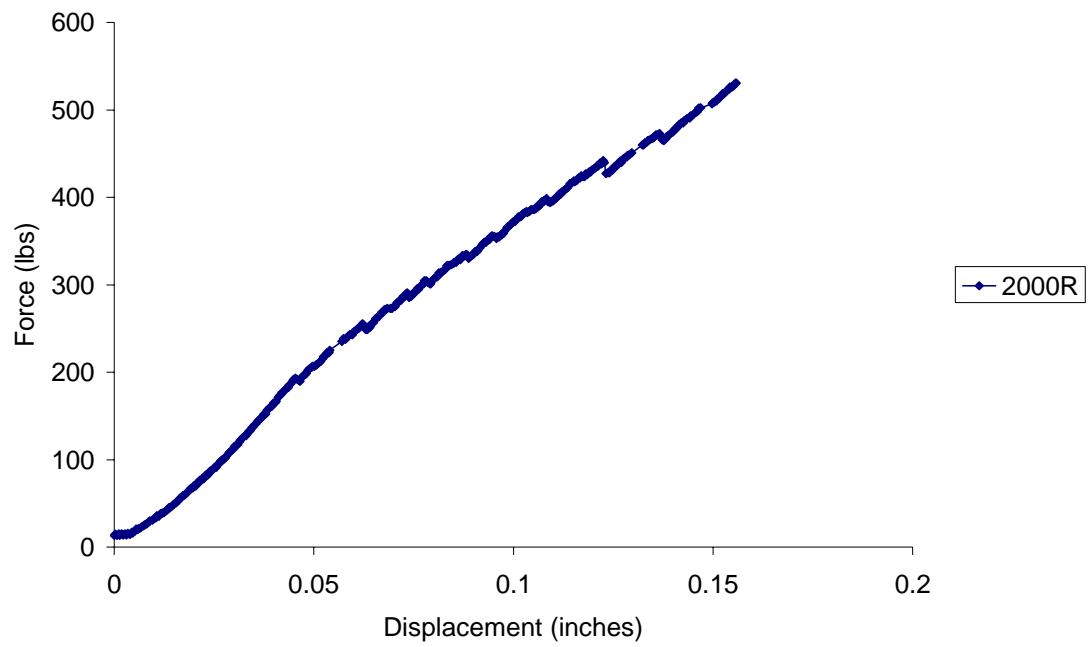
Compression Testing

Whole Femurs

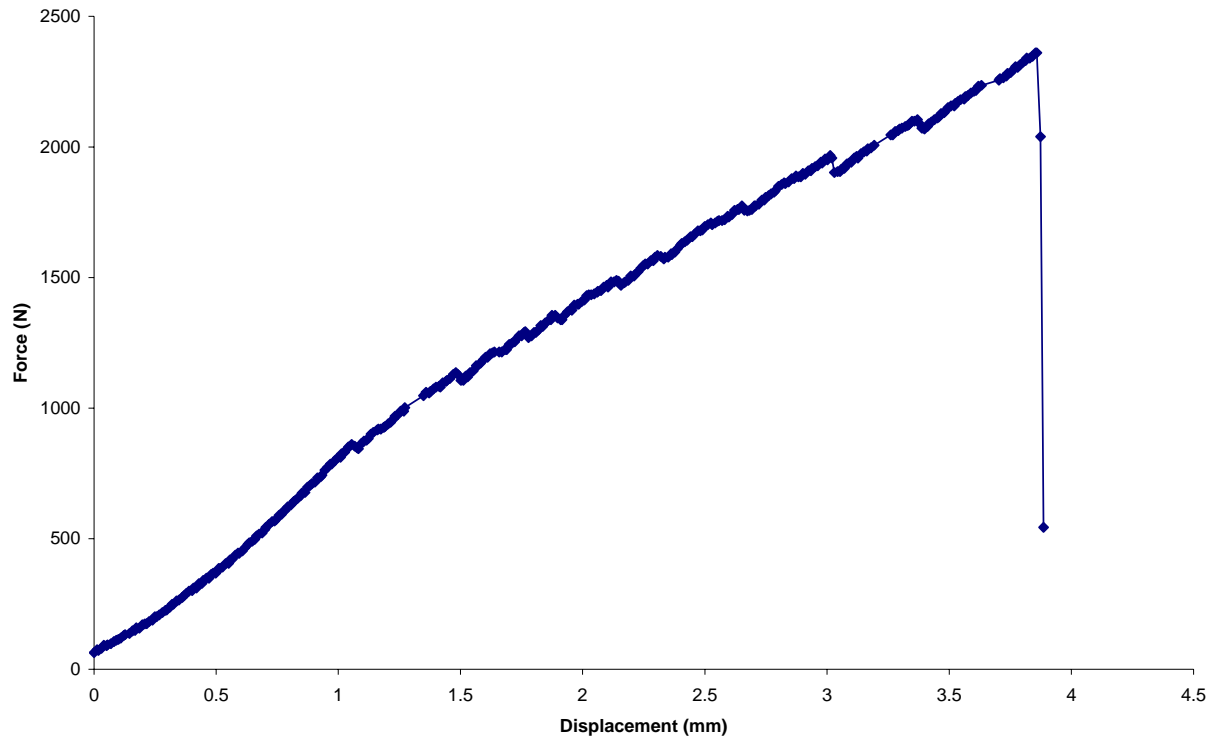


Failure of full femur

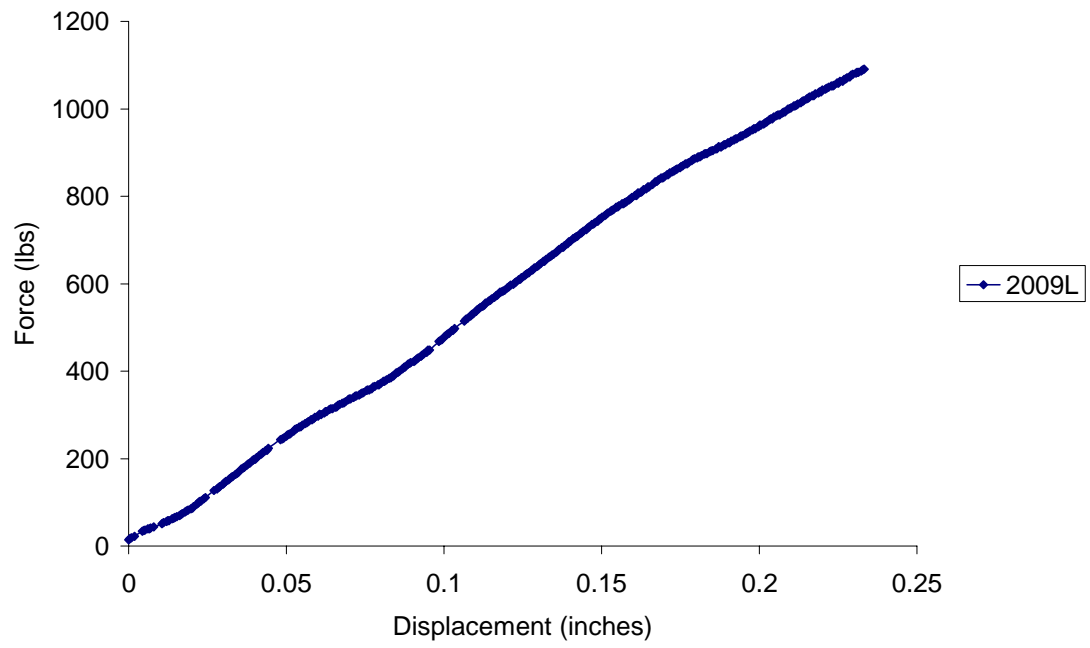
2000R



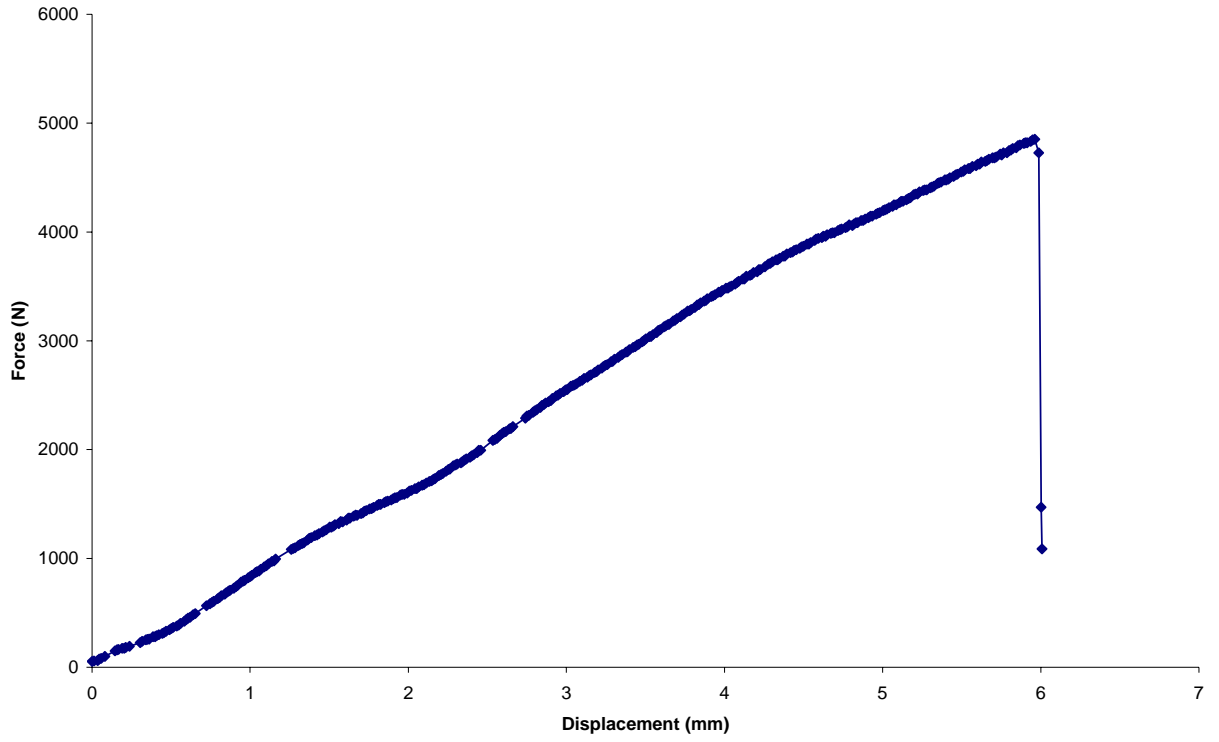
2000 R



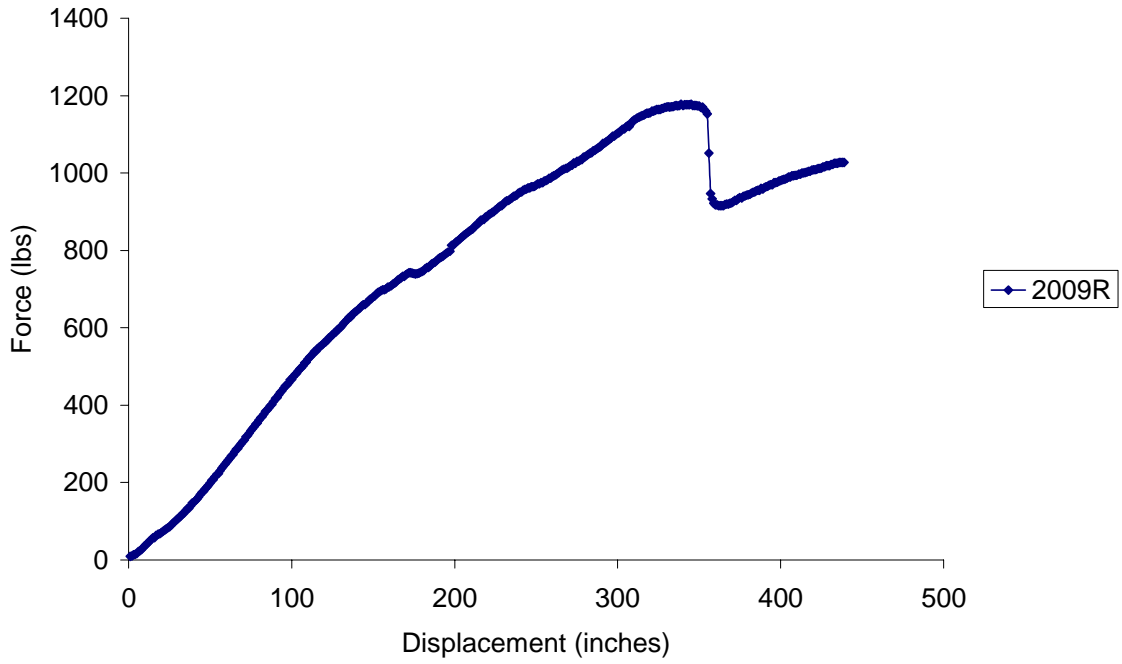
2009L



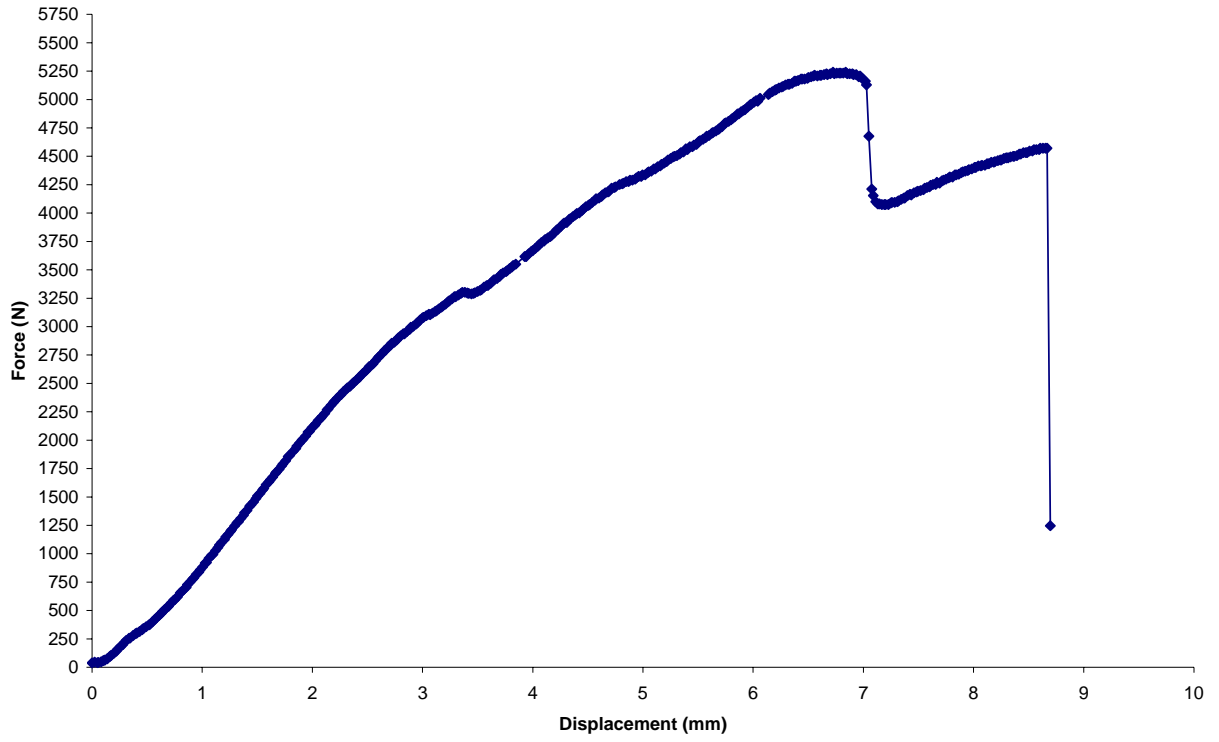
2009 L



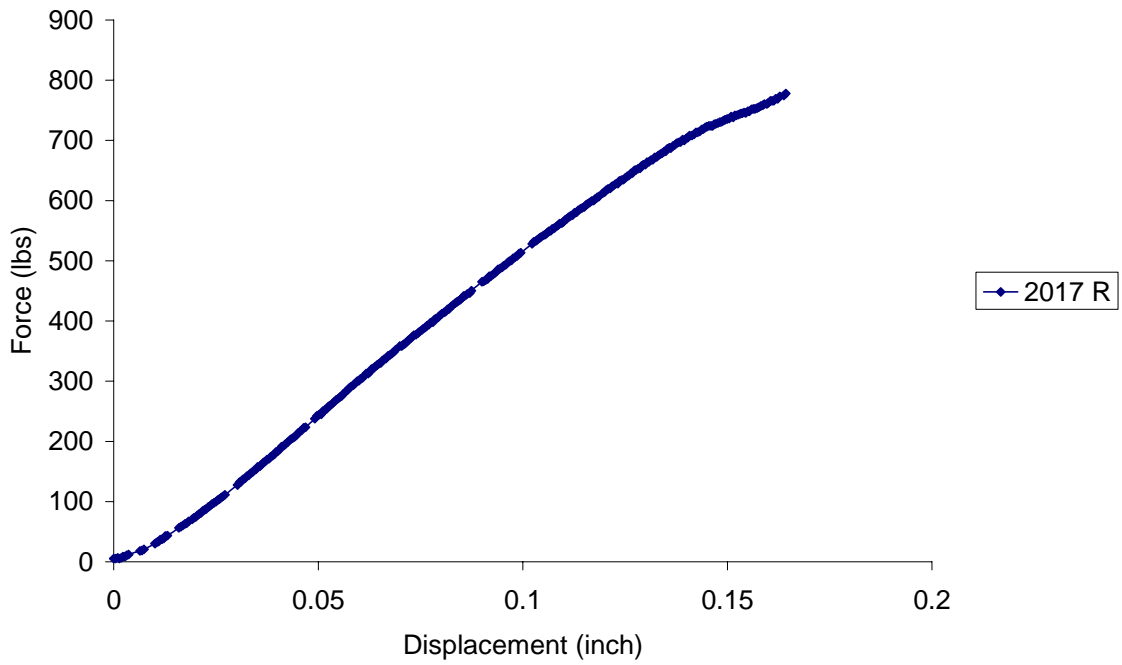
2009R



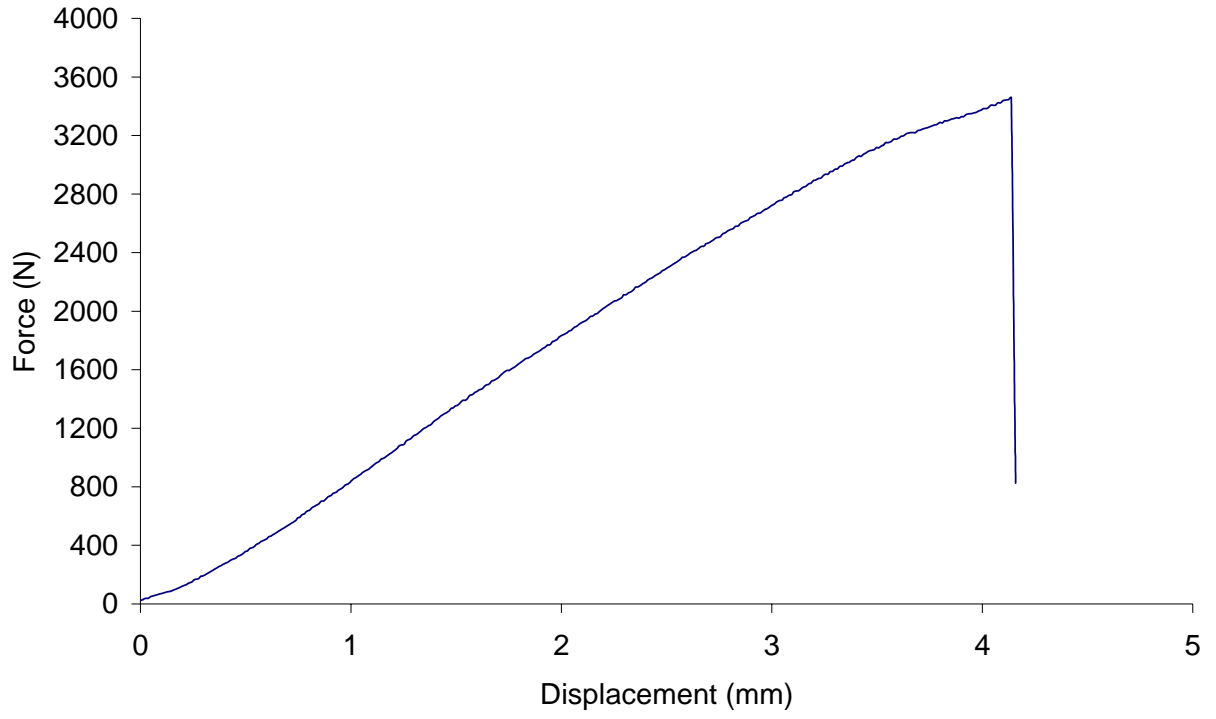
2009 R



2017 R



2017 R

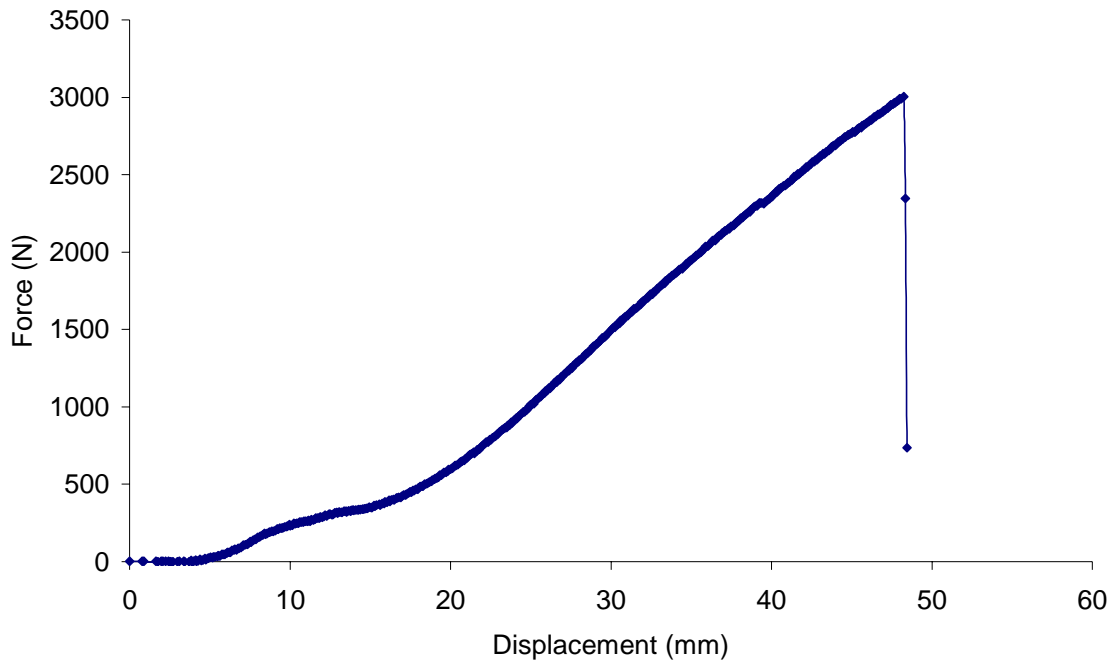


Femurs after Removal of Flap

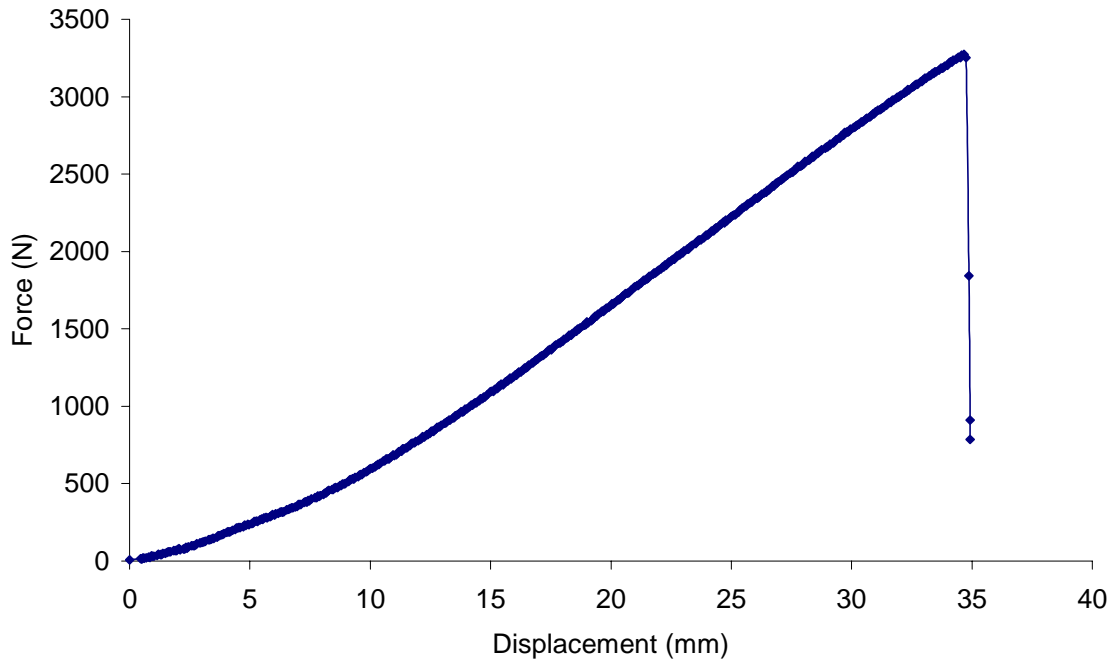


Failure of harvest femur

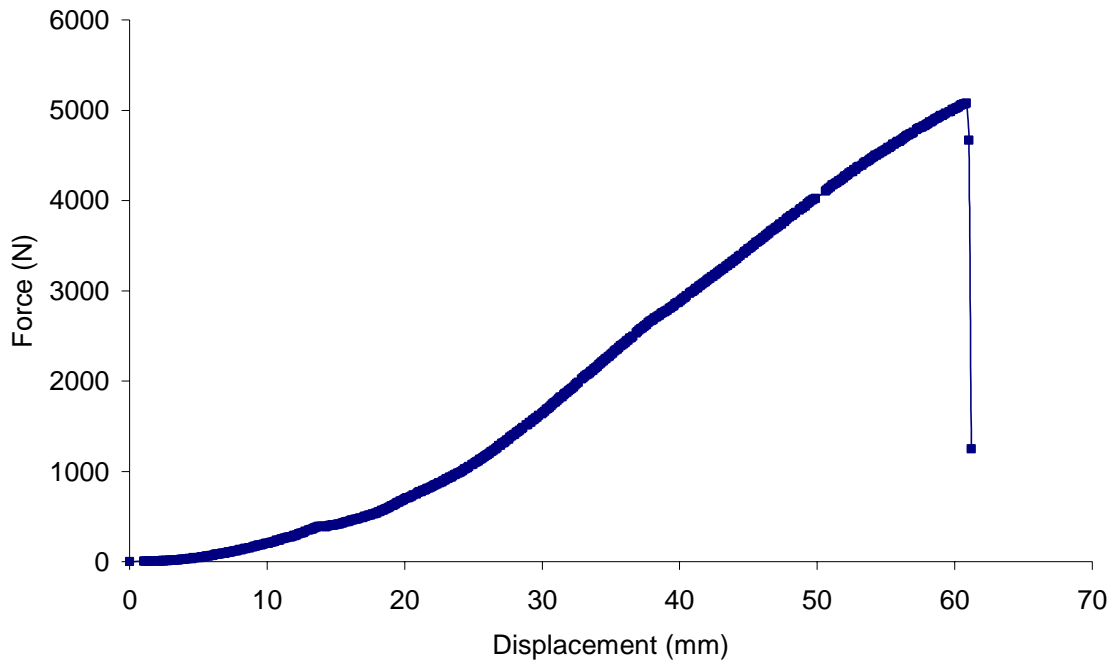
2031 L Femur (35% Removed)



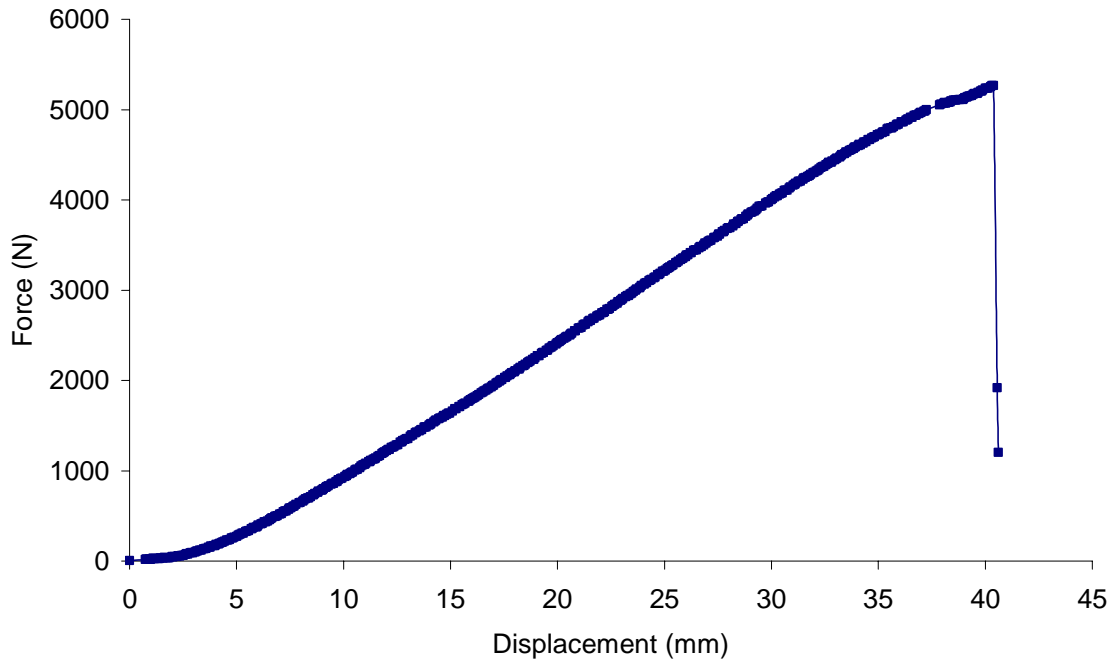
2031 R Femur (40% Removed)



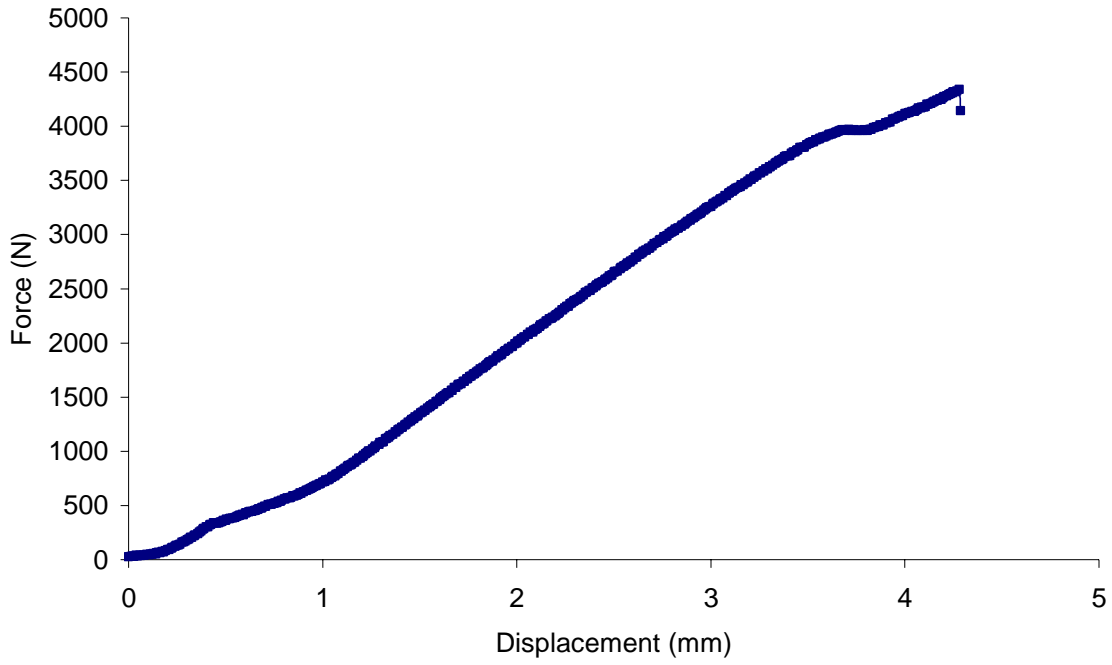
2020 L Femur (35% Removed)



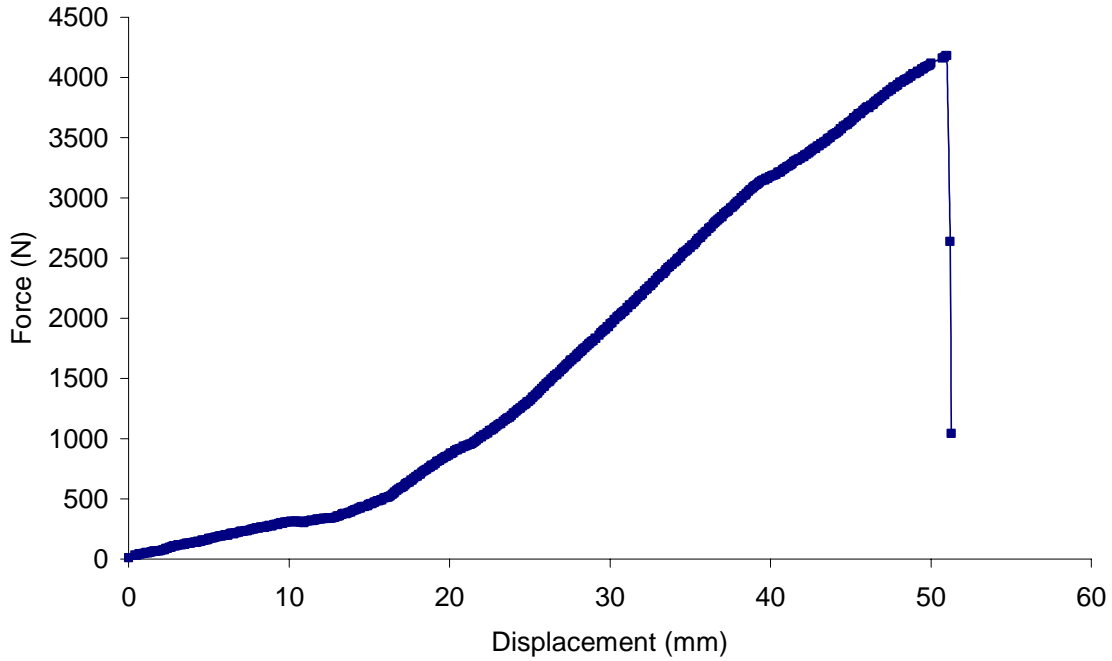
2020 R Femur (40% Removed)



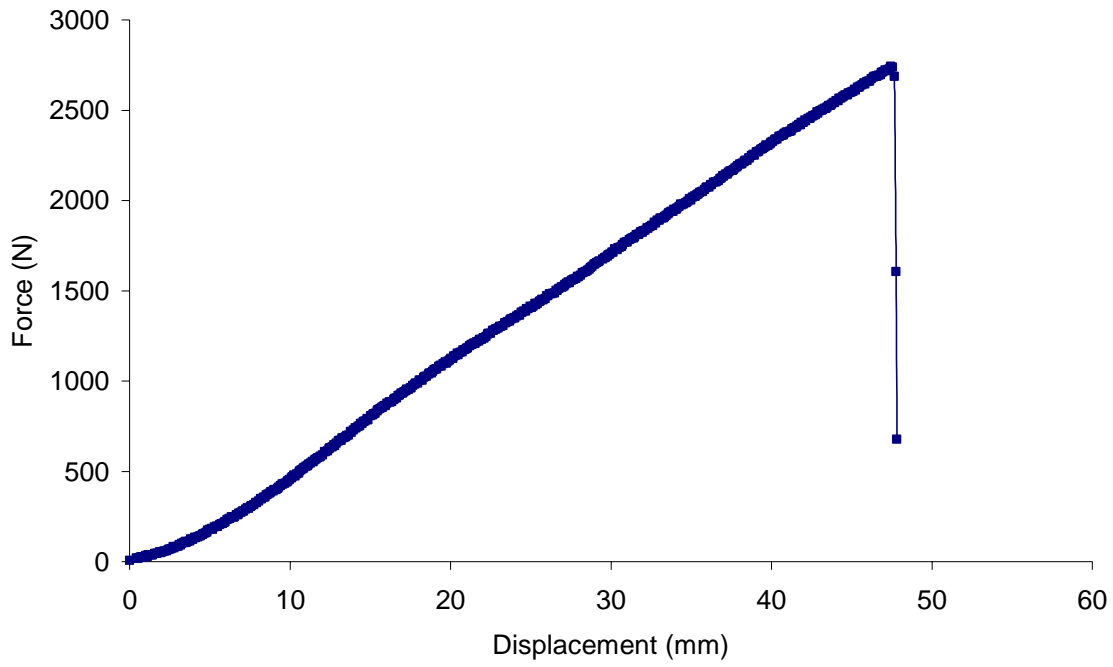
1975 L Femur (35% Removed)



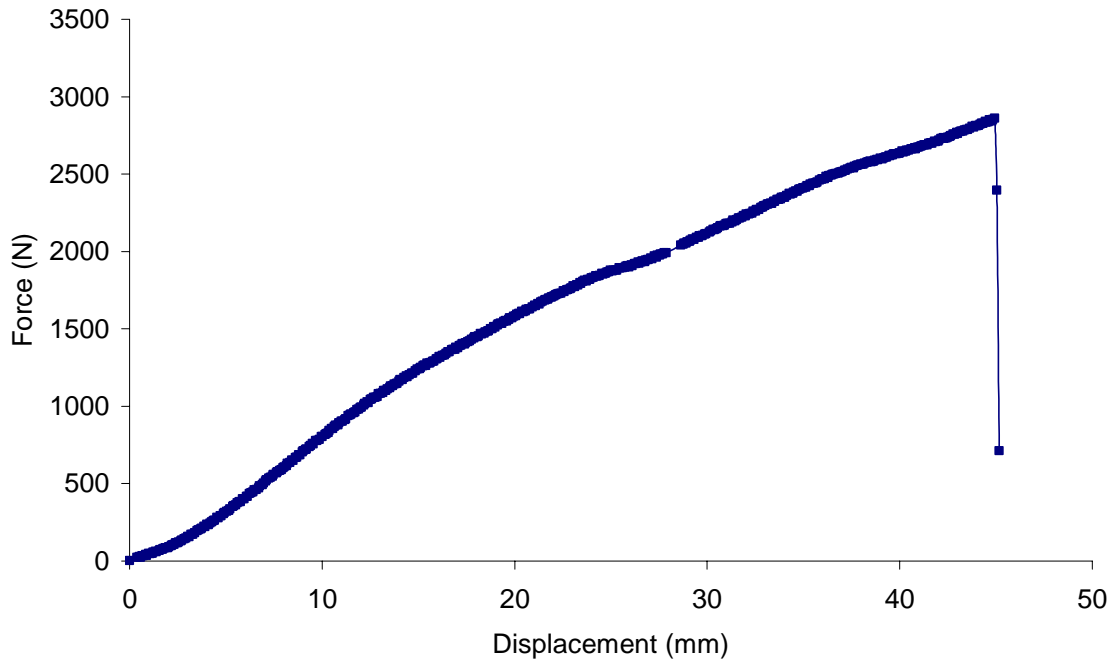
1975 R Femur (40% Removed)



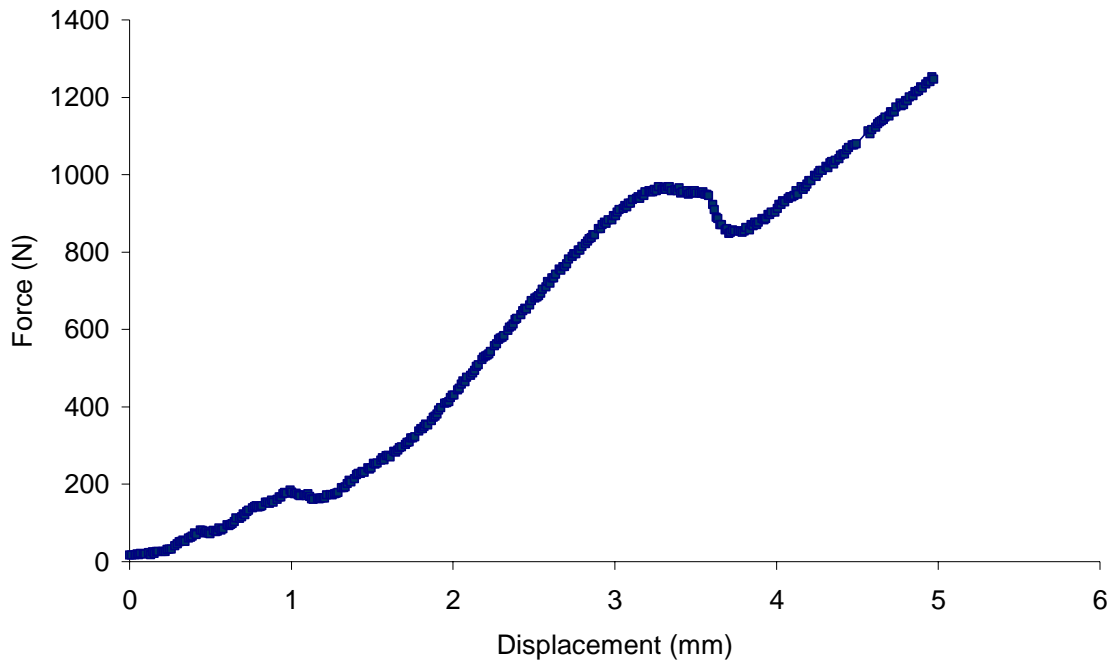
2017 L Femur (35% Removed)



2000 L Femur (35% Removed)



2028 L Femur (40% Removed)



Appendix O 3-Point Bending MTS Program Direction

(Adopted from the Test Ware-SX Running manual)

1. Enter TestStar Application Program
2. Placement of Specimen
 - a. Disable safety interlocks
 - b. Turn on the hydraulic power
 - c. For the actuator, enable the manual control
 - i. Move the actuator for placement sample
 - d. Place the sample specimen on the lower abutment of the 3-point bending attachment
 - e. Lower the actuator to a position about the sample, with no force being applied
 - f. Disable the LUC panel's control of the system
3. Zeroing
 - a. Enter into the Sensors window
 - b. Left click to select the Force Sensor
 - c. Zero the force sensor
 - i. Release the sensor's Auto Zero function
 - d. Zero the displacement sensor
4. Test Procedure
 - a. Click on TestStar Configuration
 - b. Enter the TestWare-SX application
 - c. Open a Test Template
 - i. Adjust template as need to meet the requirements of test method state in the report
 - d. Open a Test Procedure
 - i. Adjust template as need to meet the requirements of test method state in the report
5. Run Test
 - a. Enable the limits
 - i. Underpeak detection
 - ii. Error detection

- b. Begin the test
- 6. Remove of Specimen
 - a. Bring the actuator to its Home position
 - b. Turn the hydraulic pressure to low
 - c. Place the LUC to displacement mode
 - d. Remove the sample
- 7. Collect Data

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