

AFAMRL-TR-81-40

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AD A104932



**MECHANICAL PROPERTIES OF SPINAL
LIGAMENTS FOR RHESUS MONKEY, BABOON
AND CHIMPANZEE**

*ROBERT WM. LITTLE, Ph.D.
ROBERT P. HUBBARD, Ph.D.
DAVID L. HYLER*

*MICHIGAN STATE UNIVERSITY
EAST LANSING, MICHIGAN 48824*

ARNOLD R. SLONIM, Ph.D.

AIR FORCE AEROSPACE MEDICAL RESEARCH LABORATORY

JUNE 1981

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FOR THE COMMANDER



HENNING E. VON GIERKE
Director
Biodynamics and Bioengineering Division
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REPORT DOCUMENTATION PAGE		READ INSTRUCTIONS BEFORE COMPLETING FORM	
1. REPORT NUMBER AFAMRL-TR-81-401	2. GOVT ACCESSION NO. AD-A104932	3. RECIPIENT'S CATALOG NUMBER	
4. TITLE (and Subtitle) MECHANICAL PROPERTIES OF SPINAL LIGAMENTS FOR RHESUS MONKEY, BABOON AND CHIMPANZEE	5. TYPE OF REPORT & PERIOD COVERED Technical Report	6. PERFORMING ORG. REPORT NUMBER	
7. AUTHOR(s) Robert W. Little Ph.D. Robert P. Hubbard Ph.D. David L. Hyler & A.R. Slonim, Ph.D.	8. CONTRACT OR GRANT NUMBER(s) F33615-79-C-0514	9. PERFORMING ORGANIZATION NAME AND ADDRESS Michigan State University Department of Biomechanics East Lansing, Michigan 48824	10. PROGRAM ELEMENT, PROJECT, TASK AREA & WORK UNIT NUMBERS 62202F, 7231-14-09
11. CONTROLLING OFFICE NAME AND ADDRESS Air Force Aerospace Medical Research Laboratory Aerospace Medical Division, Air Force Systems Command, Wright-Patterson Air Force Base OH 45433	12. REPORT DATE June 1981	13. NUMBER OF PAGES 43	14. MONITORING AGENCY NAME & ADDRESS (if different from Controlling Office)
15. SECURITY CLASS. (of this report) unclassified		15a. DECLASSIFICATION/DOWNGRADING SCHEDULE	
16. DISTRIBUTION STATEMENT (of this Report) Approved for public release; distribution unlimited.			
17. DISTRIBUTION STATEMENT (of the abstract entered in Block 20, if different from Report)			
18. SUPPLEMENTARY NOTES			
19. KEY WORDS (Continue on reverse side if necessary and identify by block number) Spinal Ligaments Hysteresis Chimpanzee Mechanical Stress Relaxation Spinal Injuries Biomechanical Response Vertebral Levels Vibratory Loads Material Property Characteristics Rhesus Monkey Animal Models Constant Strain Rate Baboon			
20. ABSTRACT (Continue on reverse side if necessary and identify by block number) -This report covers a study of the material property characteristics of four major spinal ligaments and represents the first 1½ years of a 3-year effort; the experimental protocol to be used throughout the study is emphasized. The mechanical response characteristics of the anterior and posterior longitudinal ligaments, ligamentum flavum and supraspinous ligament were examined at different anatomical levels, and these properties were compared between the rhesus monkey, baboon and chimpanzee, three of the four species of primates to be evaluated in the total program. The following tests were conducted on these tissues:			

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establishment of initial tissue geometry, relaxation, constant strain rate, hysteresis and preliminary cyclic tests. Information on the mechanical properties of the spinal ligaments is essential to understanding the mechanism of spinal injuries that result from a constant exposure to vibratory loading environments not only during escape and crash episodes, but during routine flights especially in high performance aircraft. Different primate species data will aid in the selection of animal models and interspecies scaling techniques. ✓

SUMMARY

This report describes initial tests on primate spinal ligaments to establish their mechanical properties. These data will serve as the basis for future development of mathematical models and for comparisons between different primates. The research program is a three-year endeavor and will include tests of tissue from rhesus monkey, baboon, chimpanzee and human. This report includes data from the first three of these primates.

Four ligaments of the spine have been selected for testing and three basic tests have been performed on these ligaments. Constant strain rate loading and unloading at three different rates give basic stress-strain relations, hysteresis and strain rate effects. Cyclic testing at different frequencies give initial stress decay over short term vibration, and although these data are not reported here, they will serve as the basis for the development of long time vibration and fatigue tests. Relaxation tests give a measurement of the short term viscoelastic response while a parallel study on the stability of preconditioning gives insight into the long time flow response. These experimental results are the basis for the program's final phase, which will develop mathematical models in the form of constitutive equations to predict the response of the tissue to different stress and strain histories.

The work reported here includes the initial analysis of dynamic viscoelastic data. There are wide scatter bands of material properties but some comparisons may be made between ligaments and between the three primates. The basic stress-strain relationship can be represented by a power series and the short term relaxation by a linear function in the logarithm of time. The two time functions examined, one for short time viscous effects and the second for preconditioning stability, will serve as the basis for a nested hereditary mathematical constitutive equation. This research represents the initial investigation of the mechanical properties of spinal ligaments and is necessary to understand the mechanism of spinal injuries in aircrewmembers that result from a constant exposure to a variety of vibratory loading environments not only during escape and crash episodes, but also during the course of routine flights, especially in high performance aircraft. Differences between different primate species will aid in the selection of animal models and interspecies scaling techniques to establish criteria and develop preventive measures for the safety of aircrews.

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PREFACE

This study was conducted in the Department of Biomechanics, College of Osteopathic Medicine, Michigan State University, East Lansing, Michigan 48824, under AF Contract No. F33615-79-C-0514. Dr. Robert William Little, Professor and Chairman of the Department, was the Principal Investigator; Dr. Robert P. Hubbard was the Co-investigator. The experiments, which cover the first year and a half of a three-year effort, were conducted in support of Work Unit 72311409, "Mechanical Stress on Soft Tissue Material Properties." Dr. Arnold R. Slonim, Biodynamic Effects Branch, Biodynamics and Bioengineering Division, Air Force Aerospace Medical Research Laboratory, was the project scientist and contract monitor.

The authors gratefully acknowledge the valuable assistance of Mrs. Arvilla Bolley for her help in typing this report. The cooperation and assistance of Lt. Col. A.R. Banknieder and his staff of the Veterinary Sciences Division, Air Force Aerospace Medical Research Laboratory, and of Dr. Frederick Coulston and Dr. Charles E. Graham, Director and Deputy Director, respectively, International Center of Environmental Safety of Albany Medical College, Holloman AFB, New Mexico, in providing primate cadaveric specimens vital to this study are very much appreciated.

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Mechanical Properties of Spinal Ligaments
for Rhesus Monkey, Baboon and Chimpanzee

INTRODUCTION

The spinal column is an important mechanical system requiring study both as a system and as individual components. The physical properties of the vertebral bodies and discs have received considerable attention, but studies of the spinal ligaments have been limited in their scope. The most detailed study was done by Tkaczuk (1968) on the anterior and posterior longitudinal ligaments from seventy-four human cadaveric lumbar spines. A more recent study of these same two ligaments, but at C6 to T1, was conducted by Eddy (1977). Tkaczuk examined the load response to three cycles of deformation and yield and failure loads. Eddy limited his tests to a single constant strain rate loading and unloading.

The ligamentum flavum was studied by Nachemson and Evans (1968) and Nunley (1958). Nachemson and Evans tested ligamenta flava from the L3-L4 segment of ten human cadavers. Five load cycles were applied, followed by relaxation, cyclic tests and finally failure loadings. The ten test samples were from subjects of 13 years of age to 79 years of age and both sexes. The scatter of the data was considerable but allowed qualitative examination of the variation of ligament properties with age. The earlier work by Nunley used specimens from the cervical and lumbar regions of canines. He made comparisons between fresh, fixed and frozen specimens and found considerable differences. No provision was made to eliminate tissue dehydration either during storage or testing, and this may alter the results considerably.

A research program was initiated to study the mechanical properties of four spinal ligaments from three lower primates, rhesus monkey, baboon and chimpanzee, for later comparison with human. The anterior and posterior longitudinal ligaments, ligamentum flavum and supraspinous ligaments were taken from various spinal levels and tested in an uniaxial mode. These ligaments appear to be most important during extension and flexure.

Like many other connective tissues, spinal ligaments are structures of collagen and elastin fibers. The mechanical responses of these ligaments to extension follow the typical nonlinear response of most biological tissues (Fig. 1). One interpretation of the cause of this non-linearity is that in region I, the response is linear as elastin fibers resist extension and collagen fibers, which are wavy or not aligned with the extensional axis, are straightened or aligned but do not carry load. With further extension into region II, collagen fibers become loaded and, as they do, the tissue response stiffens. When all the collagen fibers are straightened, they resist further extension linearly in region III. In regions II and III, as in region I, the contribution of elastin is a linear resistance to extension.

The waviness of collagen fibers determines the extent of region I, the distribution of original waviness for individual collagen fibers determines the extent and shape of region II, and the quantity of collagen in the tissue determines the slope in region III. These factors of tissue structure vary among the spinal ligaments and result in differences in ligament response.

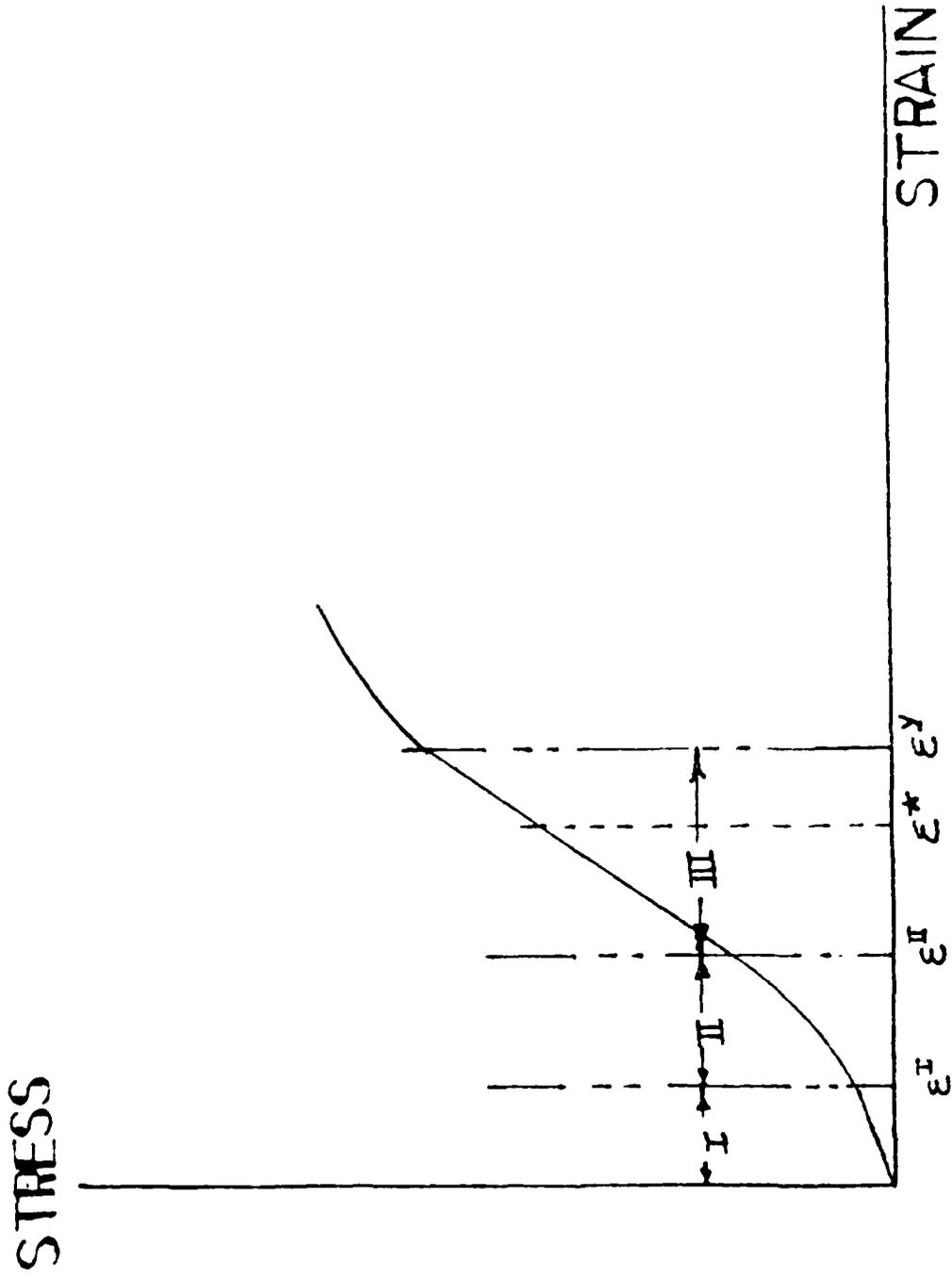


Figure 1: Typical stress-strain curve for connective tissue.

The approach in the program reported here was to repeatedly test samples at various strain rates, cyclic extensions, and relaxation. An upper limit of strain, ϵ^* , was selected in region III and the strain, ϵ^{II} , corresponding to the transition from region II to region III was defined. This permitted control of the total strain level measured relative to the transition between regions II and III.

METHODS AND MATERIALS

Many of the techniques were developed specially for this experimental research, particularly sample preparation, gripping, and testing protocol. These aspects will be discussed in some detail along with less detailed descriptions of testing equipment.

Sample Preparation:

Spinal ligament samples were all bone-ligament-bone preparations of vertebral pairs sectioned to isolate either the anterior longitudinal ligament, the posterior longitudinal ligament, the ligamentum flavum, or the supraspinous ligament. The vertebral pairs were dissected from spines that were removed intact from the test animals. Prior to dissection of the primate specimens, a procedure was developed with one embalmed human cadaver, several fresh canines, and six lower primates. These animals had been gutted with care to avoid damage to the anterior longitudinal ligament and had been subjected to a pathological examination.

The following procedure for removal of the vertebral column and sample preparation was common to all the primates used for this program. Hair was clipped from the region, then a skin incision was made from the occipital region of the head to the pelvic region, and lateral incisions were made at each end. (Fig. 2). Longitudinal incisions were made lateral to the spinous processes; the superficial, intermediate, and deep dorsal muscles of the back and neck were dissected laterally to isolate the spine, to grossly elevate the scapulae, and to expose the dorsal aspect of the rib cage. Using a Stryker saw, the ribs were cut at least 2 cm. lateral of their spinal articulations (Fig. 3).



Figure 2: Skin incision.

Figure 3: Longitudinal incisions
lateral to spinous processes



Dissecting anteriorly along the occipital bone of the skull, the vertebral column was removed from the head usually with a scalpel. Occasionally a surgical chisel was used with moderate blows from a rubber mallet. The structures on the lateral and anterior aspects of the cervical spine were severed so that the spine could be elevated from the body. Upon elevation of the cervical spine from the carcass, the cervical spine hyperextends about 90 degrees probably due to tension in the posterior ligamentous structures (Fig. 4).

Figure 4: Exposed spine.



The lumbosacral junction was identified by counting lumbar vertebrae, by relative position of the iliac crests, and by manipulation of the vertebral column with attention to the first intervertebral space that was mobile. Care was exercised to assure that sacralized and anomalous vertebrae had been identified. Sectioning of the lumbosacral junction was accomplished by cutting the supra- and intraspinous ligaments, articular facet capsule and the ligaments between the lateral lumbar processes and the pelvis. With the spine flexed, dissection continued anteriorly through the spinal nerves, disc, and ligaments of the lumbosacral junction. Often a surgical chisel was needed to free the last lumbar vertebrae from the ilia and sacrum, particularly if ossification had occurred. The anterior lumbar musculature was severed, and the vertebral column was removed from the body (Fig. 5).

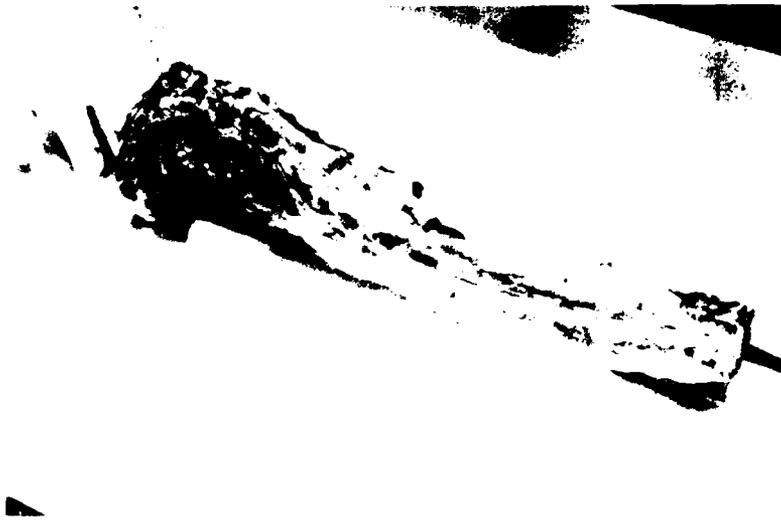


Figure 5: Excised vertebral column.

This dissection and removal of the spine could be accomplished in a few minutes, and the fascia and muscle tissues around the spine prevented noticeable tissue drying during this period. After removal, the spine was wrapped with paper towels that had been dampened with physiological saline. The spinal column was next sectioned according to the scheme shown in Figure 6 for each species.

This sectioning resulted in vertebral pairs both for mechanical testing and material for study of tissue structure using histological techniques and light microscopy and in some cases scanning electron microscopy. To section the spinal column, the intervertebral disc and surrounding ligamentous tissue were cut, the supraspinous and intraspinous ligaments and ligamentum flavum were cut, and the facet joints were disarticulated with a scalpel. For the thoracic vertebrae, the cut segments of ribs were also removed. This dissection was done

Rhesus Baboon		Chimpanzee	
Level	Test	Level	Test
C7	Mech.	C7	Mech.
T1		T1	
T2	Histo.	T2	Histo.
T3	Mech.	T3	Mech.
T4		T4	
T5	Histo.	T5	Histo.
T6	Mech.	T6	Mech.
T7		T7	
T8	Histo.	T8	Histo.
T9	Mech.	T9	Mech.
T10		T10	
T11	Histo.	T11	Mech.
T12	Mech.	T12	
L1		T13	Mech.
	Histo.	L1	
L2	Mech.	L2	Histo.
L3		L3	Mech.
L4	Histo.	L4	
L5	Mech.		
L6			
L7	Histo.		

Figure 6. Vertebral level test sectioning scheme.

under a flow of humidified air, and the vertebrae that had not been dissected were kept moist by a saline-dampened towel (Fig. 7). After sectioning, the vertebral pairs for test samples were wrapped in saline-dampened paper towels, put in airtight plastic bags, marked for identification, and frozen for storage. All the pairs from a single animal were placed in a second bag for convenient identification and as a precaution against drying while freezing.



Figure 7: Final sectioning of vertebral parts.

To prepare ligament samples for testing, the frozen vertebral pair was thawed at room temperature while wrapped in saline-dampened towels. The pair was then cut with a band-saw to section the vertebrae with three frontal planes (Fig. 8). Bone-ligament-bone preparations were then made for the ligamentous structures to be tested.

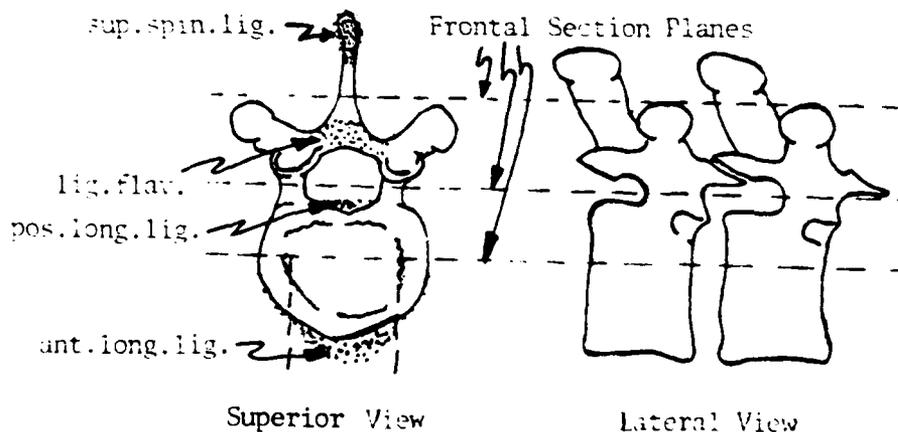


Figure 8: Saw-cuts to prepare vertebrae pair for testing.

The sample for testing the anterior longitudinal ligament (A.L.L.) was trimmed with sagittal cuts to either side of that ligament. In the rhesus and baboon, the A.L.L. is localized in the center of the vertebrae so that this trimming can be done without removing any of that ligament. In chimpanzee, particularly in the lumbar region, the A.L.L. is broader with less well defined borders so that trimming must be less extensive. Once trimmed, the inter-vertebral disc material was removed with care not to cut fibers of the A.L.L.

The posterior longitudinal ligament (P.L.L.) was prepared in much the same manner except that the lateral extent of the sample was determined by sagittal cuts at the medial edge of the pedicles for easier gripping. There are some P.L.L. fibers that course laterally at the disc space and these were cut by this sectioning. These fibers were thought to contribute little to the resistance to axial deformations.

To prepare a test sample of the ligamentum flavum (L.F.), at least one of the bony facets on each side of the ligament was removed with care to avoid damage to the L.F. The removal of the facet and destruction of the facet joint eliminated the contribution of the relatively stiff collagenous joint capsule to the mechanical response of the L.F., which is composed of predominantly elastin fibers. Occasionally some of the remnants of the spinous processes were removed to allow the L.F. to be in a state of zero tension prior to testing.

The supraspinous ligament (S.S.L.) sample was prepared from the portion of the spinous processes cut from the lamina. The intraspinal ligament was judged to be those fibers that are anterior to the S.S.L. which do not course axially between the spines and these fibers were cut. There are many tendinous fibers for muscle attachments in the posterior region of the spinous process. Complete removal of these fibers was not attempted since the S.S.L. fibers are the only ones loaded by axial movement of adjacent spinous processes. However, this functional distinction between tendinous and ligamentous fibers is not apparent histologically, and this made measurements of the cross-sectional area of the S.S.L. difficult.

Mechanical Testing Equipment:

Mechanical testing of the tissue samples was conducted with an Instron* (Fig. 9) machine that is hydraulically powered and electronically controlled to produce uniaxial extensions at rates up to 1 m/s. Sample extension (grip motion) and load were recorded with a digital storage oscilloscope and stored for subsequent analysis on flexible, magnetic diskettes. These data were transferred to a microcomputer for analysis.

Tissue samples were tested at room temperature in a chamber supplied with water-saturated air, and saline was dripped over the samples to sustain tissue moisture. Attached to the chamber base is a stereo-microscope and camera which was used to observe samples during testing. The chamber and microscope can be rotated to view the sample from different angles.

*Model 1331 (Instron Corp., Canton, MA)

Figure 9: Tissue biomechanics testing facility.



Gripping:

The spinal ligaments were sectioned from pairs of adjacent vertebrae as described and tested as bone-ligament-bone samples. The concept that the ligaments constrained the motions of adjacent vertebrae was the basis of grip design for each of the four ligament types. The bone segment and ligament attached to that bone were held by the grip to experience the axial displacements imposed by the testing machine. The grips were designed to be much stiffer than the ligament samples so that the motion of the testing machine would be equal to sample displacement.

The grips used with the anterior longitudinal ligament sample are shown schematically in Figure 10. The upper gripping plate was attached to the actuator of the testing machine and the lower plate to the load cell. The ligament sample was held against these plates with stainless steel bands. On the surfaces of both the plates and the bands, a waterproof abrasive mesh (silicon carbide 120 grit "sand screen") was epoxied to eliminate slippage without cutting the tissue. Additional gripping was provided by a screw bearing against plates in front of the gripping bands (not shown in figure).

The samples of posterior longitudinal ligament and supraspinous ligament were tested with conventional grips with abrasive mesh on their surfaces.

The ligamentum flavum samples (Fig. 11) were geometrically more complex than the other samples and did not have a large flat surface for convenient gripping. For the larger samples, the approach was to grip the sectioned pedicles and the trimmed transverse processes with threaded gripping cylinders that had annular ridges to bear against the bone (Fig. 12). These cylinders were held by yokes (not shown in the figure) which were attached to the testing machine. With the gripping cylinders in place, holes were drilled through the pedicles and

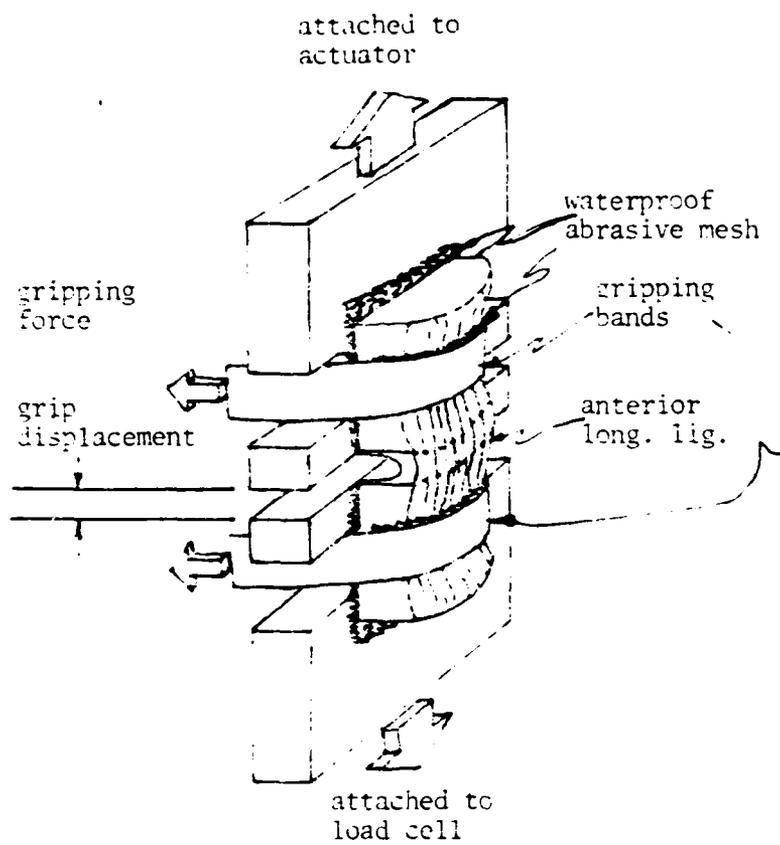


Figure 10: A.L.L. grips

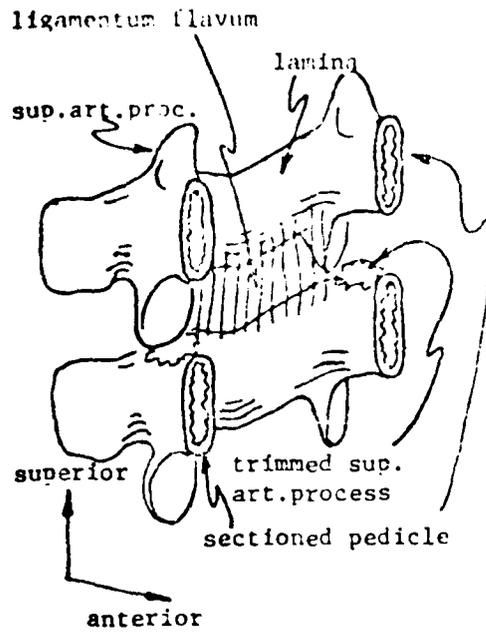
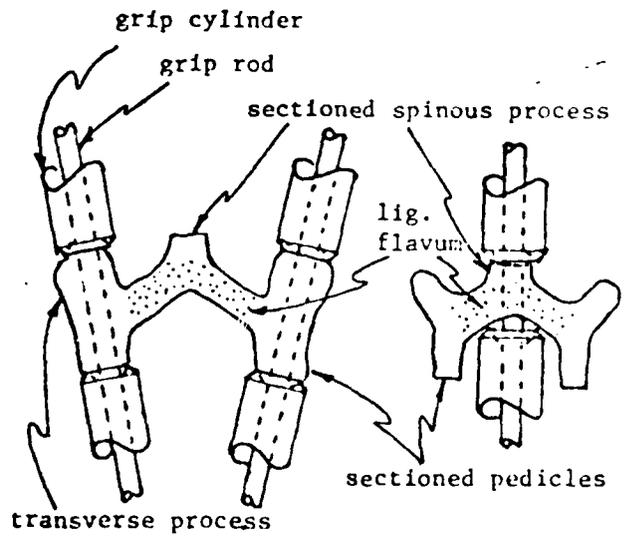


Figure 11: L.F. sample



Gripping of
larger vertebrae

Gripping of
smaller vertebrae

Figure 12: L.F. grips

transverse processes, and a rod was placed through the hole, as shown in Figure 12. In the smaller vertebrae, the pedicles were generally too narrow to be gripped or drilled so that each lamina was gripped with a single yoke at the midline. This method of gripping of the smaller samples did not eliminate the possibility of rotation about an anterior-posterior axis. However, such rotations were not observed, probably due to symmetry of the ligament about the midsagittal plane. For both the larger and smaller samples, the gripping methods allowed axial displacements to be imposed on the ligamentum flavum.

Mechanical Testing Protocol:

Preliminary tests were conducted to aid in grip refinement and to confirm the general viscoelastic nature of the tissues. The predominantly collagenous ligaments (i.e., anterior longitudinal, posterior longitudinal, and supra-spinous) exhibited load-extension responses with little or no initial linear response (region I), more abrupt stiffening in region II, and greater stiffness in region III in comparison to the samples of ligamentum flavum which were predominately elastin and responded with an extensive region I and gradual stiffening.

At the onset of the testing program, the levels of load and deflection that particular spinal ligament samples, and other structures to be studied, would sustain in in vivo activities or in in vitro testing without damage were not known. In these viscoelastic tissues, small extension will cause structural changes that will tend to revert with time, but above some extension level structural changes will occur, such as collagen fiber rupture, that are irreversible in vitro and would require healing in vivo to restore normal function.

The repeatability of response to various levels of extension within region III (Fig. 1) was studied and led to the criterion for selection of ϵ^* as an extension in region III which typically was associated with a load of twice the load at the transition between region II and III (ϵ^{II}). Thus, ϵ^* was the maximum extension for any particular sample and was selected during the first extension of the sample.

Preliminary testing also focused on the repeatability of response to successive extensions, the relaxation of load, and the load response to haversine extensions at various frequencies. With this information, a protocol was established and is listed below:

A. PRECONDITIONING

- 1) Thaw test sample wet with normal saline.
- 2) Mount sample and tighten grips.
- 3) Ramp slowly to establish a strain level ϵ^* well into linear region III, which will be the maximum non-destructive strain.
- 4) Hold at ϵ^* for 2 minutes and tighten grips.
- 5) Unload and wait 10 minutes.
- 6) Ten constant rate cycles of 1% per second to ϵ^* .
- 7) Wait 5 minutes.
- 8) Determine initial unloaded length ℓ_0 .
- 9) Three tests to ϵ^* at 1% per second with 5 minute wait after each test.

B. CONSTANT STRAIN RATE LOADING AND UNLOADING

- 1) One test at 100% per second to ϵ^* followed by 5 minute wait.
- 2) One test at 1% per second to ϵ^* followed by 5 minute wait.
- 3) One test at 0.1% per second to ϵ^* followed by 5 minute wait.
- 4) Two tests to ϵ^* at 1% per second with 5 minute wait after each to check preconditioning stability.

C. CYCLIC TESTS

Examination of the data from the last test in B.4 will establish the strain, ϵ^{II} , at the transition from the non-linear toe region, region II, to the linear region III.

- 1) Cyclic strain from $0.4\epsilon^{II}$ to ϵ^{II} at 10 Hertz for 40 seconds followed by a 5 minute wait.
- 2) Using the same minimum and maximum strains as test C.1, cycle 40 seconds at 1 Hertz followed by a 5 minute wait.
- 3) Using same minimum and maximum strains as test C.1, cycle 40 seconds at 0.1 Hertz followed by a 5 minute wait.
- 4) Check preconditioning stability by test B.4.
- 5) Using a strain equal to $\epsilon^{II} + 0.2(\epsilon^* - \epsilon^{II})$ as a minimum and ϵ^* as the maximum level, cycle 40 seconds at 10 Hertz followed by a 5 minute wait.
- 6) Using the minimum and maximum strains from C.5, cycle 40 seconds at 1 Hertz followed by a 5 minute wait.
- 7) Using the minimum and maximum strains from C.5, cycle 40 seconds at 0.1 Hertz followed by a 5 minute wait.
- 8) Check preconditioning stability by test B.4.

D. RELAXATION

From the second test in C.8, determine new ϵ^{II} transition strain or confirm ϵ^{II} from test series C.

- 1) Ramp at 100% per second to $0.7\epsilon^{II}$ and hold until relaxation approaches zero (approximately 10 minutes).
- 2) Return to zero strain and wait an equal time as relaxation time in D.1.
- 3) Ramp at 100% per second to ϵ^* and hold until relaxation approaches zero (approximately 25 minutes).
- 4) Return to zero strain and wait an equal time as relaxation time in D.3.
- 5) Check preconditioning stability by test B.4.

Geometric Properties:

Initial length of a ligament was defined as the smallest bone-to-bone distance of the ligament and was measured with the unloaded sample in place in the testing machine. Rough dimensions of ligament cross section were also taken at this time, but areas used in data analysis were obtained by measurements from histological slides made after testing. The initial lengths and areas of the different ligaments are given in Table 1.

TABLE 1: GEOMETRIC PROPERTIES OF LIGAMENTS

Ligament	Level	RHESUS		BABOON		CHIMPANZEE	
		A x 10 ⁻⁶ m ²	ℓ ₀ x 10 ⁻³ m	A	ℓ ₀	A	ℓ ₀
S.S.L.	C7 - T1					3.09	5.1
	T3 - T4			0.511	4.6	1.76	4.83
	T6 - T7	0.948	5.38	1.50	3.7	4.18	6.4
	T8 - T9					6.12	3.6
	T9 - T10			1.84	2.3		
	T11 - T12					12.63	2.3
	T12 - L1	1.18	5.38	3.62	5.79	24.3	
	L2 - L3			5.10	5.1		
	L3 - L4	0.815	7.11	5.84	10.4	18.31	
	L5 - L6	0.859	7.01	3.16	14.2		
L.F.	C7 - T1					27.8	4.15
	T3 - T4			34.66	4.4	29.4	4.1
	T6 - T7	29.88	5.1	49.46	4.9	27.2	5.2
	T8 - T9					34.4	5.0
	T9 - T10	36.54	5.8	43.64	2.3		
	T11 - T12					50.2	8.2
	T12 - L1	54.85	8.5				
	L2 - L3			92.95	5.1		
	L3 - L4					70.7	9.5
	L5 - L6	53.31	1.6	120.75	11.2		
P.L.L.	C7 - T1					8.28	2.5
	T3 - T4			1.29	2.2	2.94	3.7
	T6 - T7	0.752	1.9	3.28	1.8	5.48	2.0
	T8 - T9	1.04	2.7			3.42	2.8
	T9 - T10	1.04	3.1				
	T12 - L1	1.167	3.8				
	L2 - L3			3.07	3.8		
	L3 - L4	0.933	3.4				
	L5 - L6	0.908	2.8	3.65	6.0		
		0.985	3.5	1.06	5.7		
A.L.L.	T3 - T4			11.53	3.9	58.54	2.5
	T6 - T7	5.75	2.3			43.9	2.0
	T8 - T9	6.23	4.4			65.4	3.9
	T9 - T10	8.99	3.2	25.58	3.6		
	T11 - T12					46.32	5.3
	T12 - L1	9.59	4.1	31.62	3.7		
	L2 - L3			24.0	2.8		
				36.8	3.5		
	L3 - L4	8.99	3.4				
	L5 - L6	9.95	3.6	38.19	5.1		
	7.79	3.9	37.50	6.9			

RESULTS

All data were collected in digital form with the oscilloscope and transferred to the computer for analysis. The constant strain rate loading and unloading data were smoothed to eliminate noise by use of a least square polynomial expansion numerical technique. These expansions were used to calculate stress-strain data, tangent moduli, energy input, recovery and hysteresis. The hysteresis effect was measured both by the actual energy dissipated and by the percent of input energy dissipated.

Typical constant strain rate test data are shown in Figure 13; note the strain rate effects and the hysteresis area for highest strain rate. The fastest strain rate results for each ligament and animal species are shown in Table 2. These results are summarized in Table 3 and indicate a large scatter in the data. Much of the scatter may be due to large variations in strain rates. There is not a significant difference between species in the supraspinous ligament data, but differences do exist in the other ligaments. The ligamenta flava in the chimpanzee are stiffer than in the other two primates, and these ligaments are stiffer in the upper vertebral levels.

Values of tangent modulus measured are in the lower range of those reported in the literature for collagenous connective tissue, primarily tendon. This range is from $30-800 \times 10^6 \text{ N/m}^2$, but low values might be expected for spinal ligaments due to the non-parallel fiber morphology of these tissues.

The large strains which these tissues were able to withstand again may be expected due to fiber orientation and the deformation before fibers were aligned and loaded. Strain values are highly dependent upon definition of initial length of the ligament. Individual fibers have different points of attachment and in some cases span several vertebral bodies. Initial length was defined as the minimum distance between the bony surfaces in the bone-ligament-bone sample and subject to some inconsistency. However, any definition of original length would still yield higher strains than observed in parallel fibered tendons. The hysteresis data cannot be easily compared to other connective tissues as few viscoelastic tests of this nature have been conducted.

Two types of relaxation have been examined, and at this time it is not understood if they are independent or not. The first is the short-recovery time, viscoelastic relaxation which was tested by standard relaxation tests. The results are shown in Figures 14 to 17. The normalized relaxation function may be approximated as a linear function of the logarithm of time.

$$G(t) = 1 - \mu \ln(t+1)$$

The relaxation coefficient μ gives a measure of the tissue viscoelasticity. This coefficient varied from 3.0 to 11.0 for the supraspinous ligament, which had the most scatter, and from 1.0 to 3.0 for the least viscoelastic tissue, ligamentum flavum. The high elastin content of the latter leads one to expect reduced viscous effects.

The second time dependent effect was measured by the stability of the preconditioning check loop. Preconditioning of tissue prior to testing has become an accepted but not understood procedure. Upon initial loading, tissue exhibits

TABLE 2: MECHANICAL DATA AT MAXIMUM CONSTANT STRAIN RATE.

Ligament	Species	Spinal Level	Strain Rate 1/sec	Strain %	Stress N/m ² x 10 ³	Input Energy N/m ² x 10 ³	Hysteresis Energy N/m ² x 10 ³	Hysteresis % of Input Energy	Tangent Modulus N/m ² x 10 ³	
S.S.L.	Chimpanzee	C7-T1	105.8	43.2	143	15.09	2.01	13.3	108.96	
		T3-T4	119	54.2	333	40.24	8.47	21.1	221.91	
		T6-T7	165	41.9	119	8.21	0.99	12.1	124.8	
		T8-T9	234	88.1	156.3	24.6	4.02	16.3	74.26	
		T11-T12	406	49.4	158.4	33.7	6.34	18.8	52.5	
	Baboon	T3-T4	157	53.5	660	69.96	12.13	17.3	479	
		T6-T7	176	51.5	119	11.1	1.8	15.2	102.5	
		T12-L1	184	15.7	18.8	0.99	0.20	20.0	30.1	
		L2-L3	124	35.5	90.5	4.83	0.49	10.1	111.6	
		L5-L6	281	52.1	73.3	11.7	2.22	19.01	38.8	
	Rhesus	T12-L1	114	42.7	109.3	8.56	1.59	18.6	107.4	
		L5-L6	215	23.9	34.6	2.52	1.19	47.2	41.1	
	L.F.	Chimpanzee	C7-T1	180	26.2	17.8	1.20	0.058	4.86	23.2
			T3-T4	245	48.7	55.7	5.87	0.309	5.25	43.5
			T6-T7	463	43.9	50.9	5.68	0.131	2.30	48.5
T11-T12			100	26.1	23.3	1.75	0.328	18.7	27.8	
Baboon		T6-T7	100	34.5	7.25	1.11	0.17	15.3	4.86	
		T9-T10	681	100	33.7	2.07	0.186	8.99	9.43	
		L2-L3	458	81.6	11.9	3.15	0.273	8.67	3.83	
		L4-L5	284	41.7	11.85	1.58	0.108	6.82	7.695	
		L5-L6	100	29.5	3.81	0.438	0.069	15.78	2.58	
Rhesus		T6-T7	130	24.5	27.1	2.38	0.23	9.64	25.41	
		T9-T10	104	81.8	18.5	3.43	0.64	18.55	7.66	
		T12-L1	104	15.3	1.14	0.09	0.026	28.7	1.22	
		L3-L4	100	7.31	2.12	0.086	0.024	28.6	4.26	
		L5-L6	1000	186	28.0	12.63	1.76	13.9	5.26	
P.L.L.		Chimpanzee	C7-T1	238	26.6	54.6	4.58	0.835	18.26	56.44
	T3-T4		121	28.2	309.5	22.0	5.05	22.36	358.1	
	T6-T7		468	47.7	167.0	24.0	1.58	6.59	103.2	
	T8-T9		0.94	39.7	182.2	18.4	5.56	30.32	143.2	
	Baboon	T3-T4	1.0	33.9	315.2	35.1	9.88	28.2	189.2	
		T6-T7	290.8	36.8	113.5	15.8	2.59	16.45	58.1	
		L5-L6	108.1	38.8	148.1	14.6	3.18	21.82	120.7	
		L5-L6	145	31.5	335.4	28.2	6.98	24.7	315.8	
	Rhesus	T6-T7	172	32.4	135.4	12.8	2.71	21.2	91.8	
		T12-L1	182	33.2	175.1	20.2	5.67	28.1	98.3	
		L3-L4	192	36.4	88.9	7.52	3.05	40.4	86.8	
		L5-L6	108	44.0	505.0	94.1	26.4	28.1	174.6	
	A.L.L.	Chimpanzee	T3-T4	628	35.8	27.1	3.31	1.43	10.0	17.7
			T6-T7	535	50.3	23.2	3.39	0.32	9.42	13.4
			T8-T9	359	37.4	27.4	4.07	0.70	17.1	10.4
T11-T12			274	48.4	17.8	8.77	2.44	28.9	16.9	
Baboon		T3-T4	0.92	29.8	38.6	4.08	1.17	28.7	25.4	
		T9-T10	431	37.0	42.4	6.68	1.24	18.5	16.1	
		T12-L1	465	46.5	31.1	5.48	0.86	15.8	9.96	
		L2-L3	100	49.5	14.0	2.05	0.53	25.7	7.22	
		L5-L6	107	27.2	23.6	1.72	0.39	22.5	26.8	
Rhesus		T12-L1	469	21.8	28.3	1.32	0.36	27.0	54.0	
		L3-L4	4.44	25.9	72.3	6.17	1.49	24.5	64.7	
		L5-L6	100	58.8	87.5	10.3	2.58	25.1	61.0	

TABLE 5: SUMMARY OF MECHANICAL PROPERTIES

Ligament	Species	Tangent Modulus MPa	% Hysteresis	Max Strain %	Max Stress MPa
S.S.L.	Chimpanzee	52 - 220	12 - 21	41 - 88	12 - 55
	Baboon	30 - 480	10 - 20	16 - 53	9 - 66
	Rhesus	41 - 107	18 - 47	24 - 45	5 - 11
L.F.	Chimpanzee	23 - 49	2 - 19	26 - 49	2 - 6
	Baboon	2 - 9	7 - 16	30 - 100	1 - 3
	Rhesus	1 - 25	10 - 29	7 - 186	.1 - 3
P.L.L.	Chimpanzee	56 - 358	7 - 30	27 - 48	5 - 31
	Baboon	58 - 316	16 - 28	31 - 39	11 - 34
	Rhesus	87 - 175	21 - 40	32 - 44	8 - 50
A.L.L.	Chimpanzee	10 - 18	9 - 29	36 - 50	2 - 5
	Baboon	7 - 27	16 - 29	27 - 50	1.5 - 4
	Rhesus	54 - 61	24 - 27	22 - 59	3 - 9

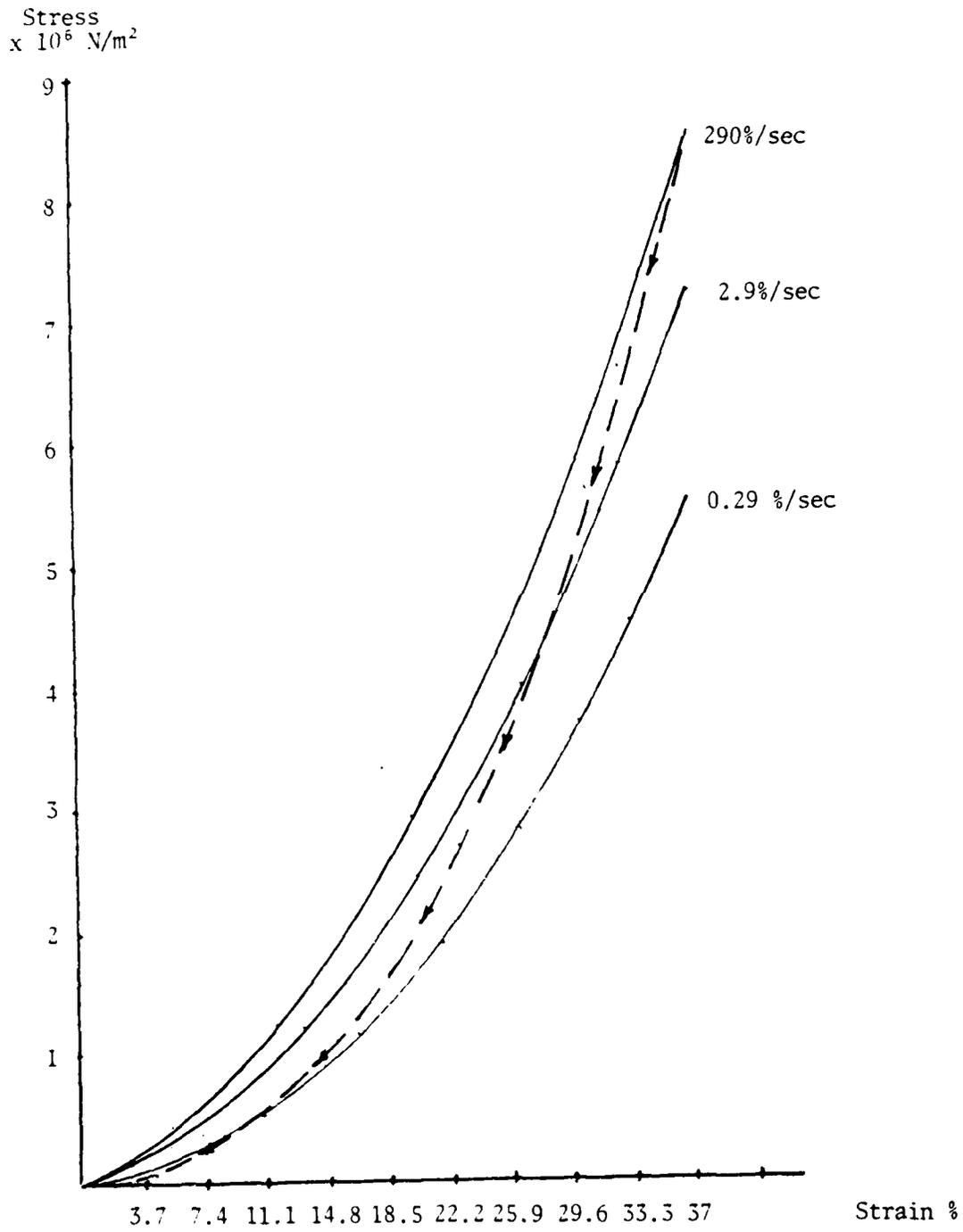


Figure 13: Typical constant strain rate data (P.L.L.-T6-T7 Baboon).

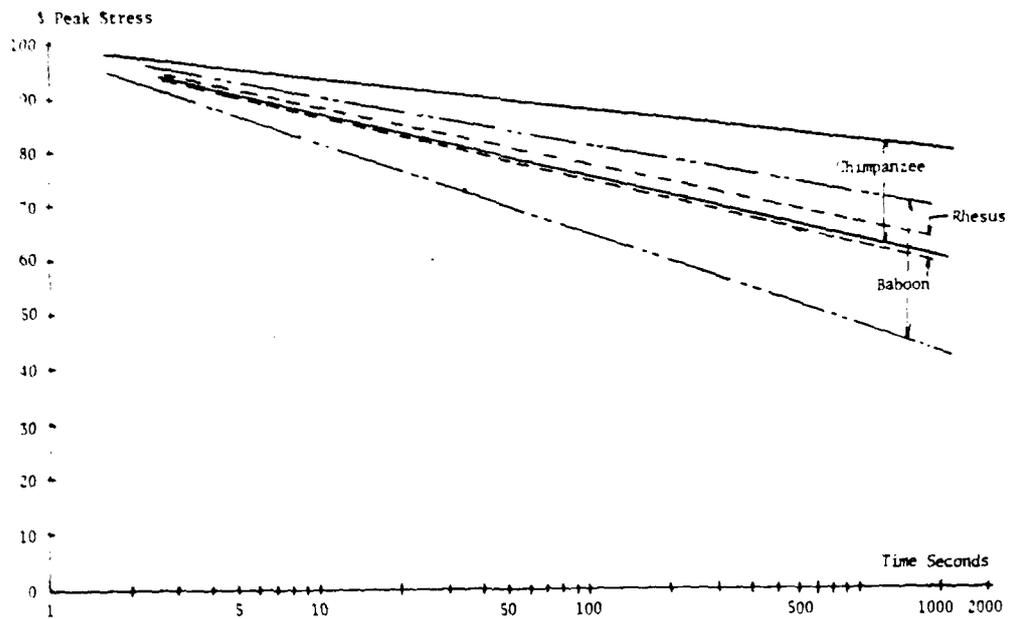


Figure 14: Viscoelastic relaxation of S.S.L.

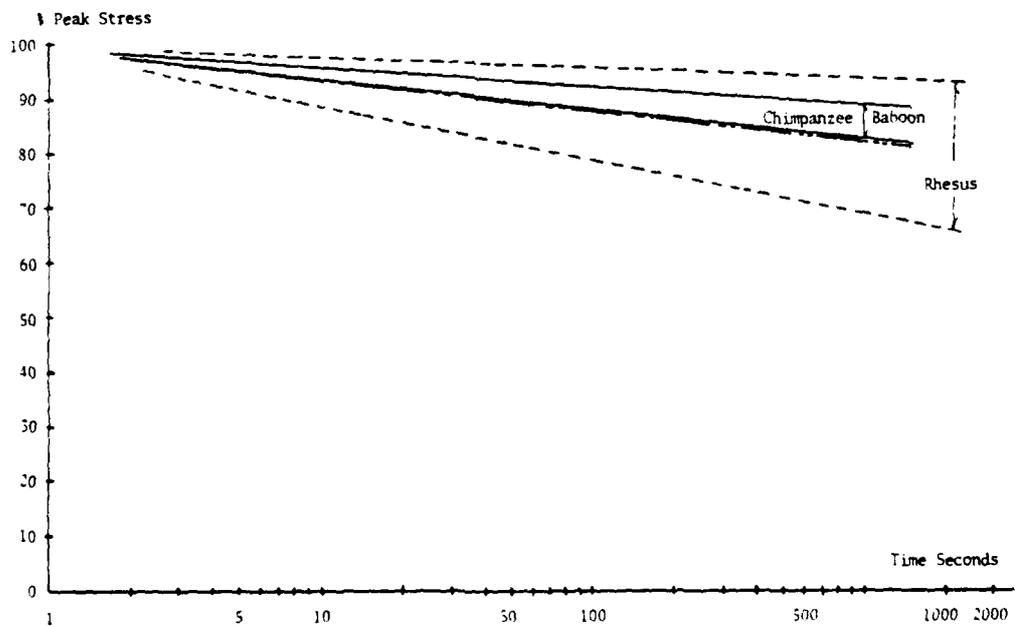


Figure 15: Viscoelastic relaxation of L.F.

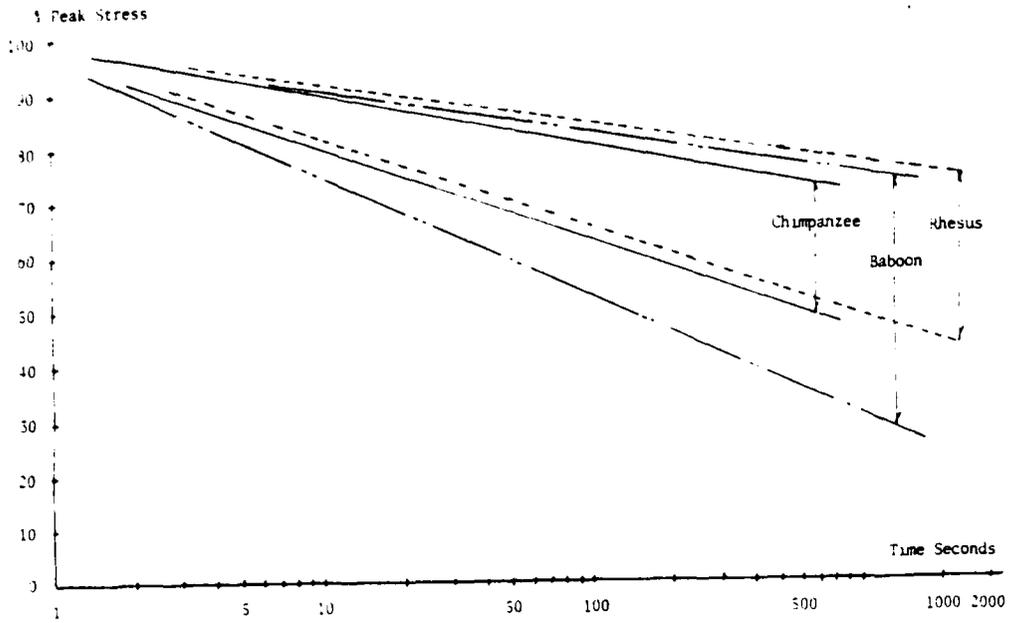


Figure 16: Viscoelastic relaxation of P.L.L.

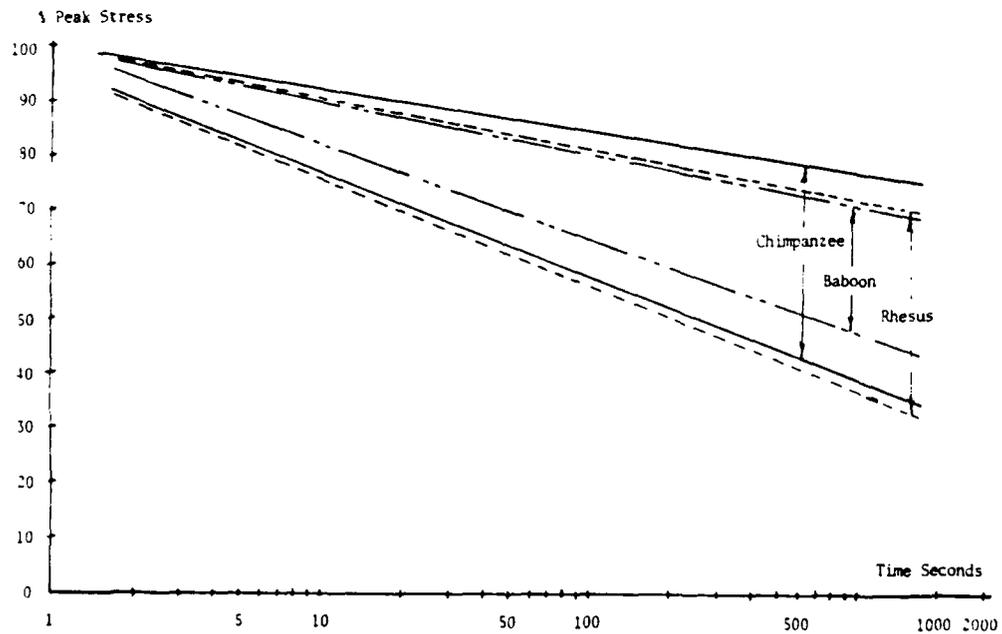


Figure 17: Viscoelastic relaxation of A.L.L.

marked changes in mechanical properties making test results non-reproducible. To stabilize the test procedure, the tissue is preconditioned by subjecting it to repeated tests at levels of stress or strain equal to or above those to be used in the test program. It is not known whether the changes which occur are independent of the other observed viscoelastic effects or not. It has been suggested that these preconditioning effects are due to changes in the state of cross-linking, alteration in the state of hydration, or realignment of the fiber matrix. As listed in the experimental protocol, ten constant strain rate tests at 1% per second to maximum test strain were used to precondition the tissue. During this procedure the stress level dropped to 85-90 percent of that attained on the initial cycle (Table 4). Subsequently, check loops were run after each test to examine the stability of the preconditioning.

The results of these checks are shown in Figures 18 to 21. The ligamentum flavum shows the most stable preconditioning, and in general ligaments from lower vertebral levels were most stable. This variation in preconditioning stability makes comparison of different tests in a testing sequence difficult as tissue response is altered by preconditioning state.

The viscoelastic and preconditioning phenomena were also examined by recording strain rate effects upon maximum stress and stiffness and upon hysteresis. Figures 22 to 25 show, for each ligament, the change in stress and stiffness with decreasing strain rate. These changes are normalized as percent of values at maximum strain rate. Both stiffness and stress decrease with decrease in strain rates as the rates drop to 1% and 0.1% of the highest rate. The last data for a 1% of maximum rate test are taken from the final preconditioning check loop and show large scatter due to the combined effects of both viscoelastic and preconditioning flows. These data emphasized the importance of a highly controlled testing protocol as identical tests run at different times in the experimental testing sequence will yield different results due to long term preconditioning effects. Recovery times of these two phenomena have not been established but appear to be 5 to 10 minutes for the short time effect and hours for the preconditioning.

The examination of hysteresis dependency upon strain rate and preconditioning gave the greatest scatter in data both for vertebral levels and for different primates. Although hysteresis was measured as the energy dissipated and as a percent of input energy, no pattern could be established except for a general trend for hysteresis to be greater for lower strain rates than at maximum rate. These results are shown in Figures 26 to 29.

Any predictive model for spinal ligaments will have to include two separate time effects, one which will have a time history over the entire test life of the tissue and a second which may be initiated for each individual subtest.

CONCLUSIONS AND FUTURE DIRECTIONS

The work reported here includes the initial analysis of the viscoelastic properties of the spinal ligaments and provides preliminary information needed to develop mathematical models in the form of constitutive equations. The "elastic" response is in the form of a power function, and there appears to be two time responses requiring a nested integral type of hereditary integral. Preliminary cyclic data have been recorded but analysis will await continued vibration

TABLE 4: TYPICAL PRECONDITIONING RESPONSE

	PRECONDITIONING (% of Peak Stress)		
	1st	Cycle 5th	10th
L.F. (T12-L1)	100	95.7	93.1
(L3-L4)	100	97	95
(L5-L6)	100	97	94
(T6-T7)	100	93	87
(C7-T1)	100	94	92
S.S.L. (L5-L6)	100	91	86
(T11-T12)	100	96	94
(T3-T4)	100	88	83
P.L.L. (C7-T1)	100	96	93
(T6-T7)	100	93	89

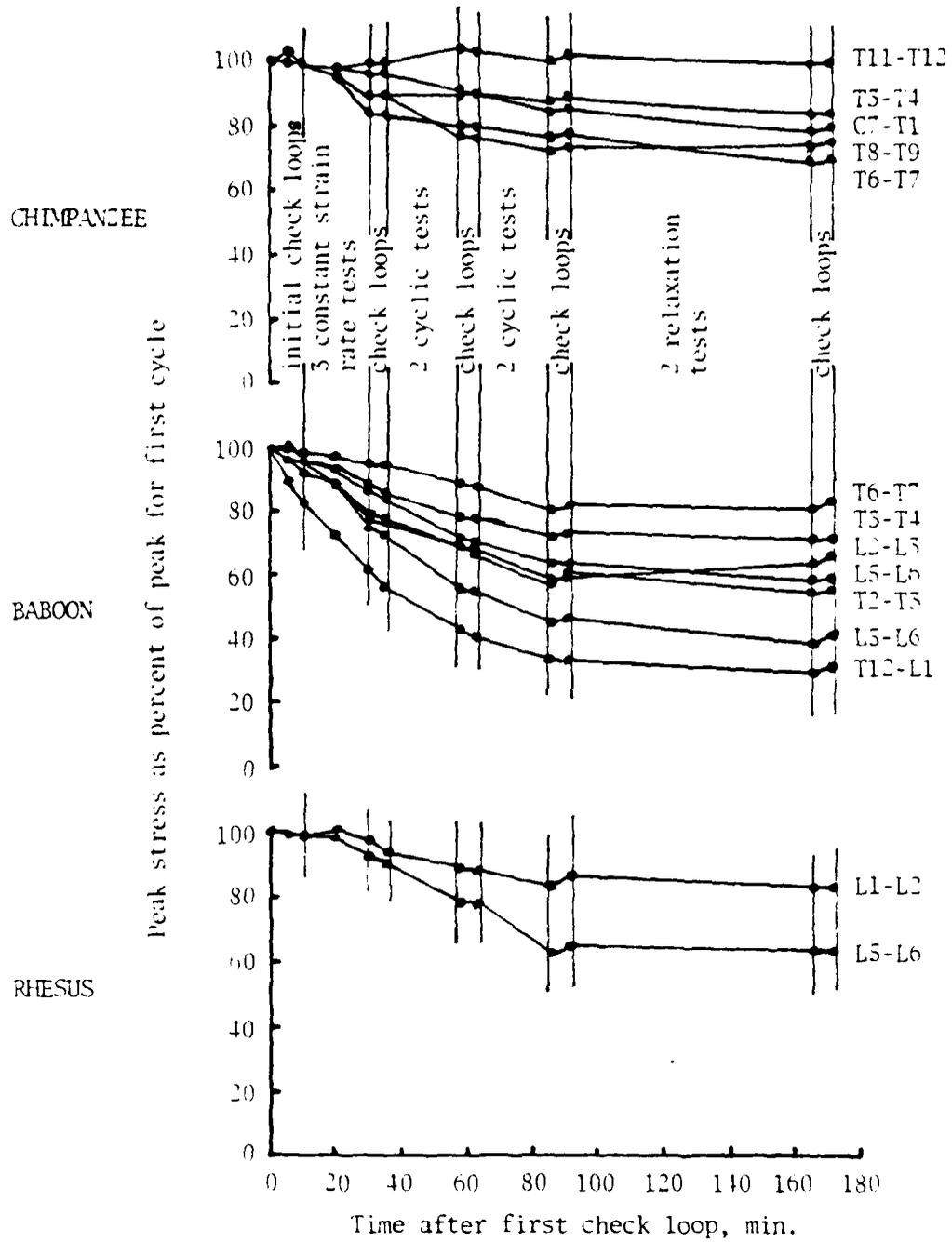


Figure 18: Preconditioning stability of S.S.L.

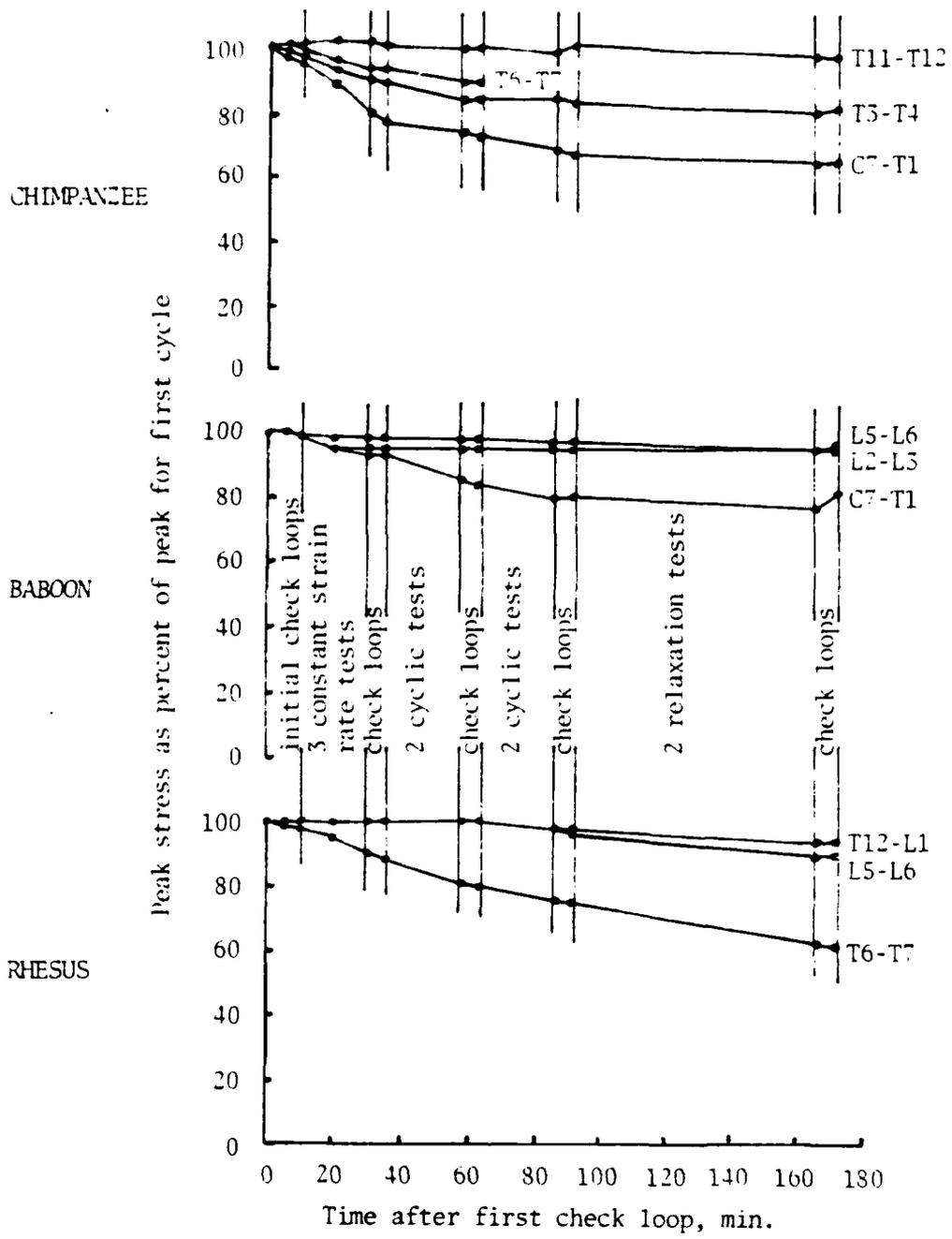


Figure 19: Preconditioning stability of L.F.

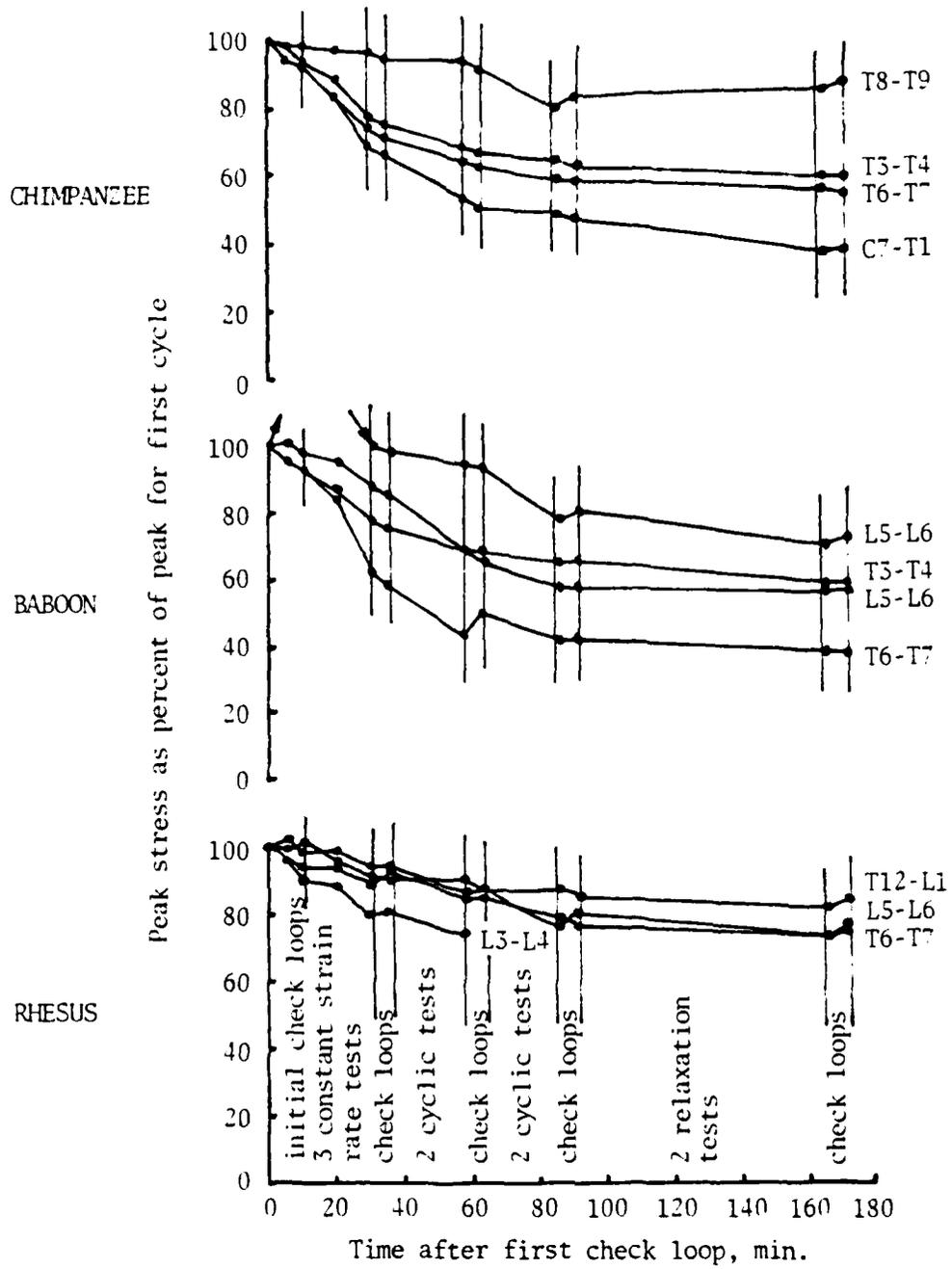


Figure 20: Preconditioning stability of P.L.L.

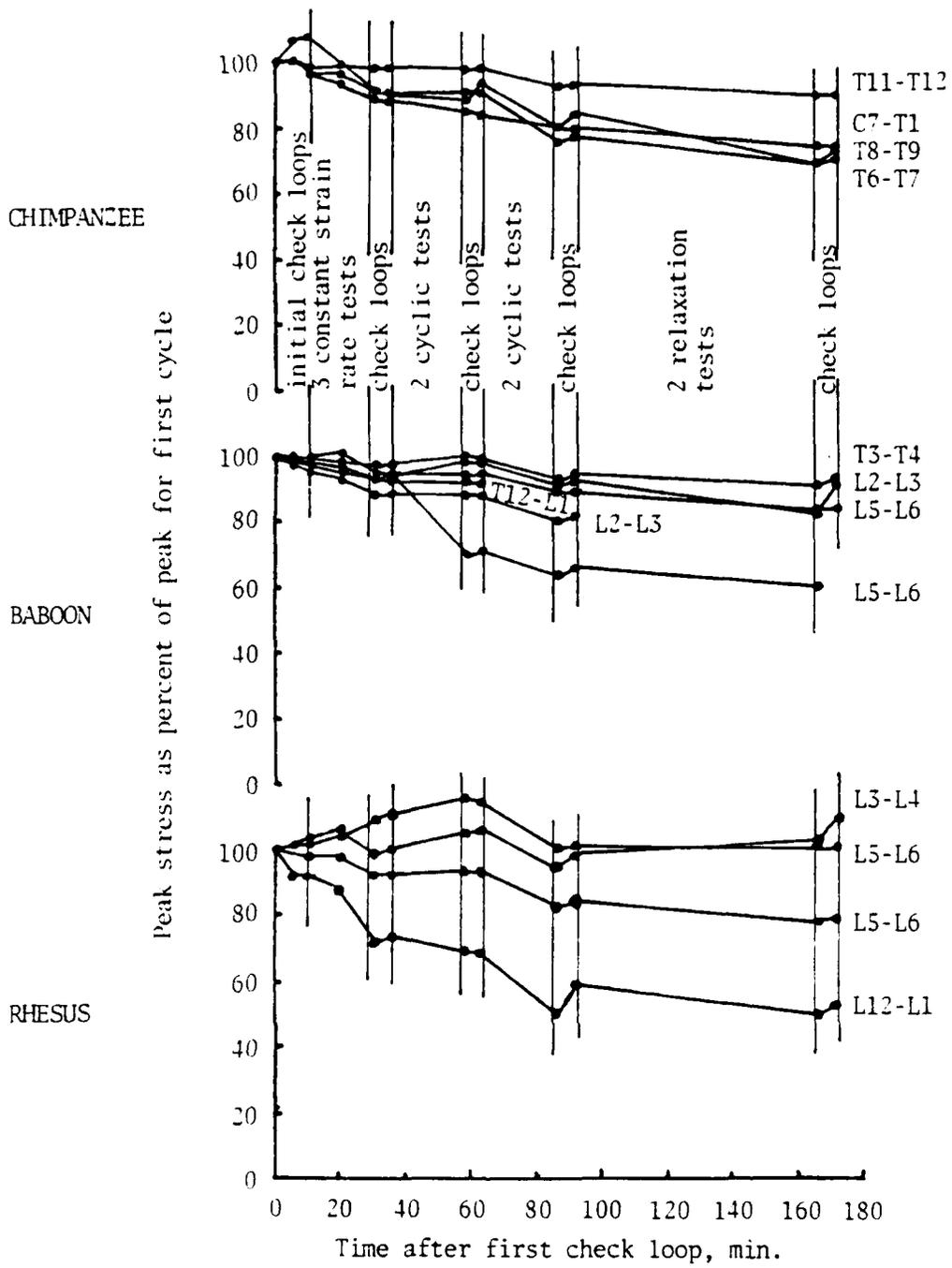


Figure 21: Preconditioning stability of A.L.L.

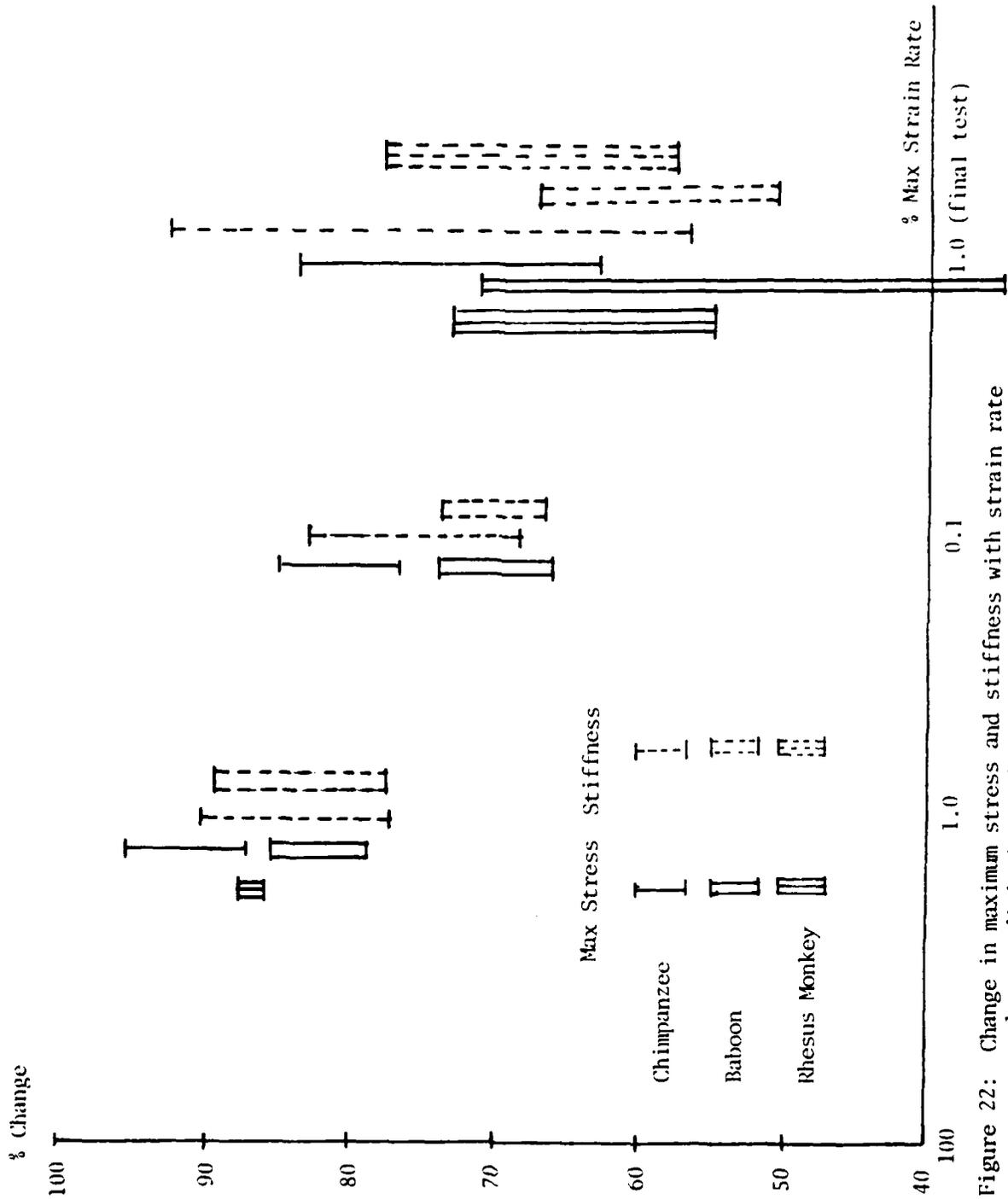


Figure 22: Change in maximum stress and stiffness with strain rate and preconditioning stability for S.S.I.

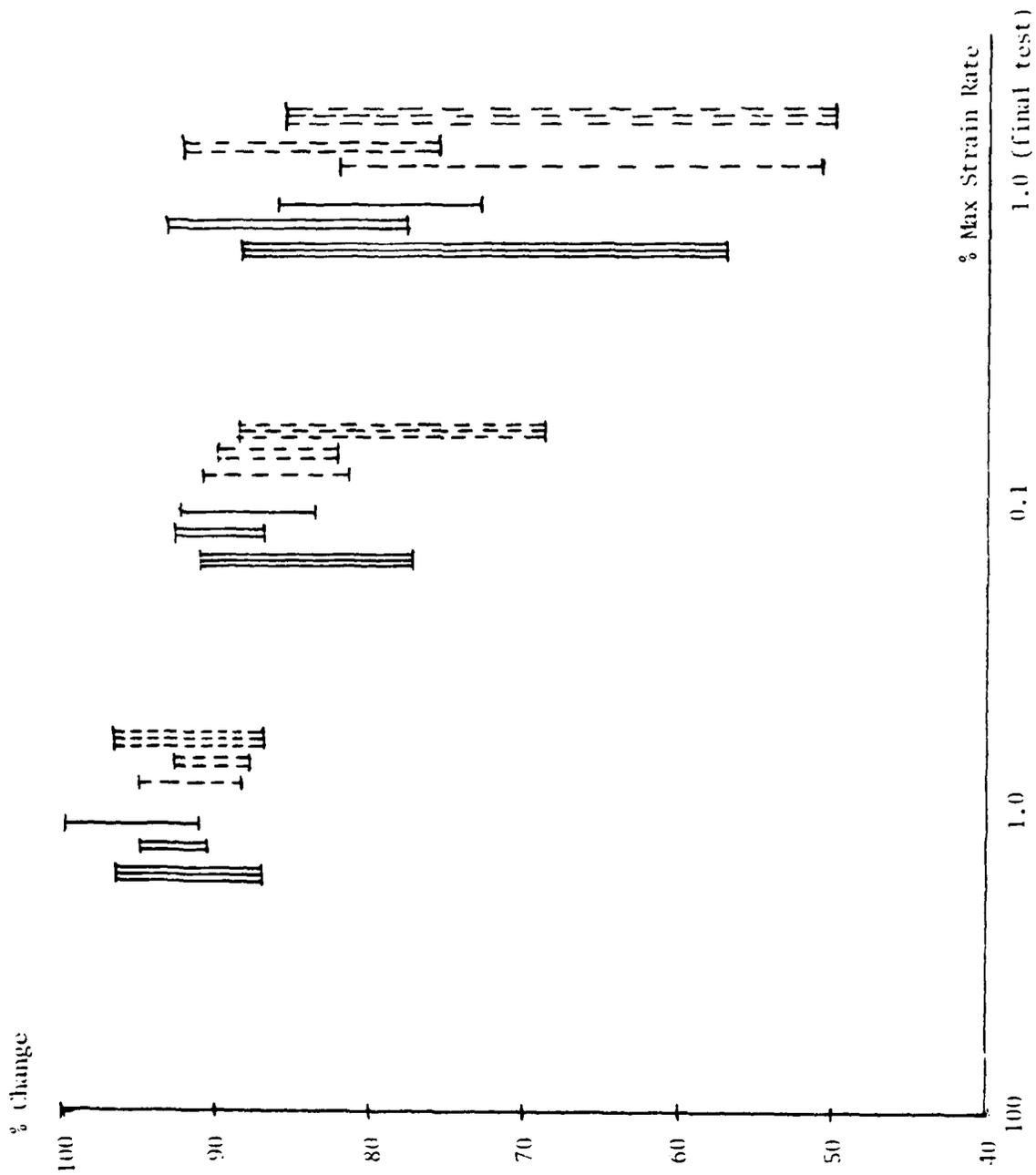


Figure 23: Change in maximum stress and stiffness with strain rate and preconditioning stability for I.F.

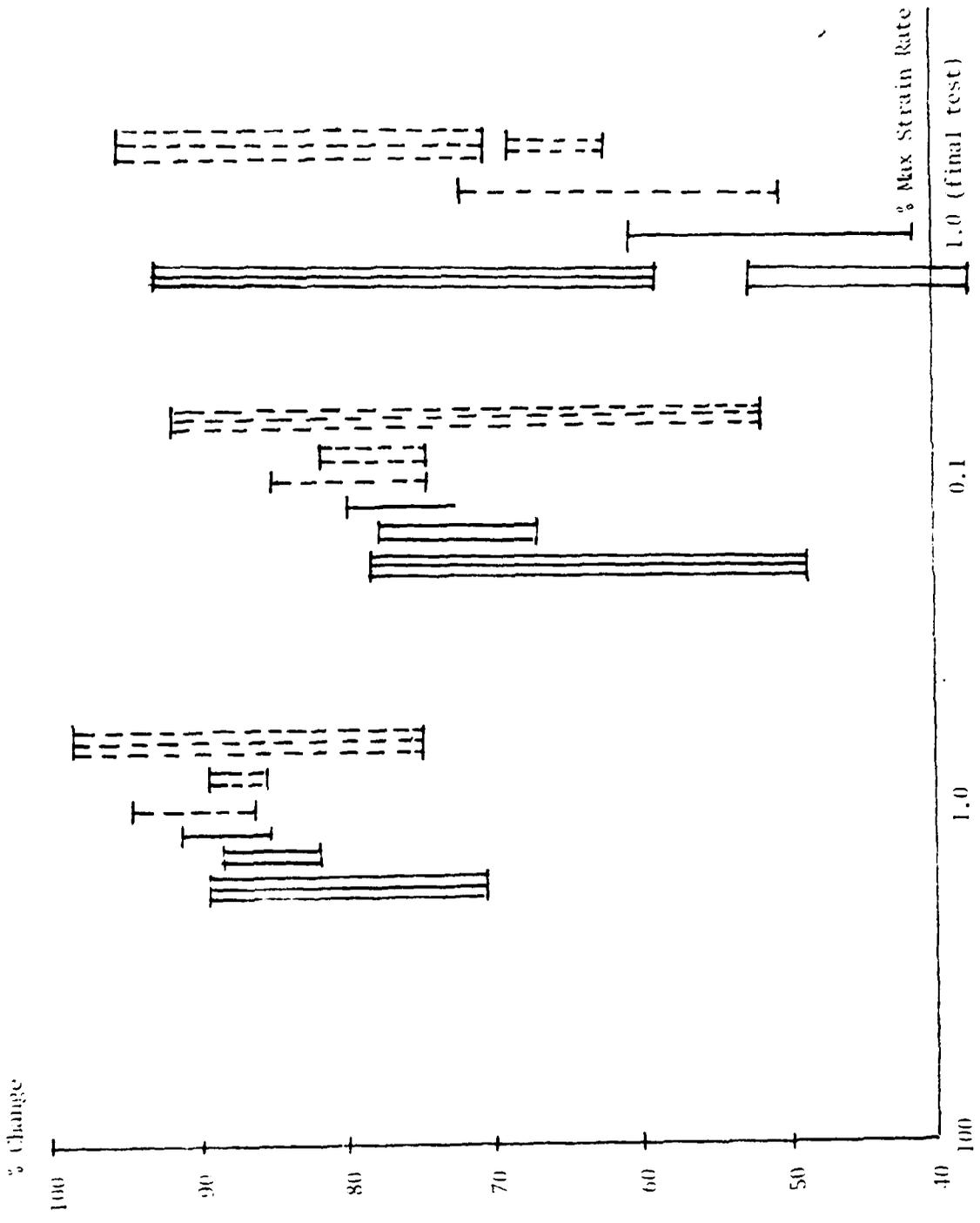


Figure 24: Change in maximum stress and stiffness with strain rate and preconditioning stability for P.L.I..

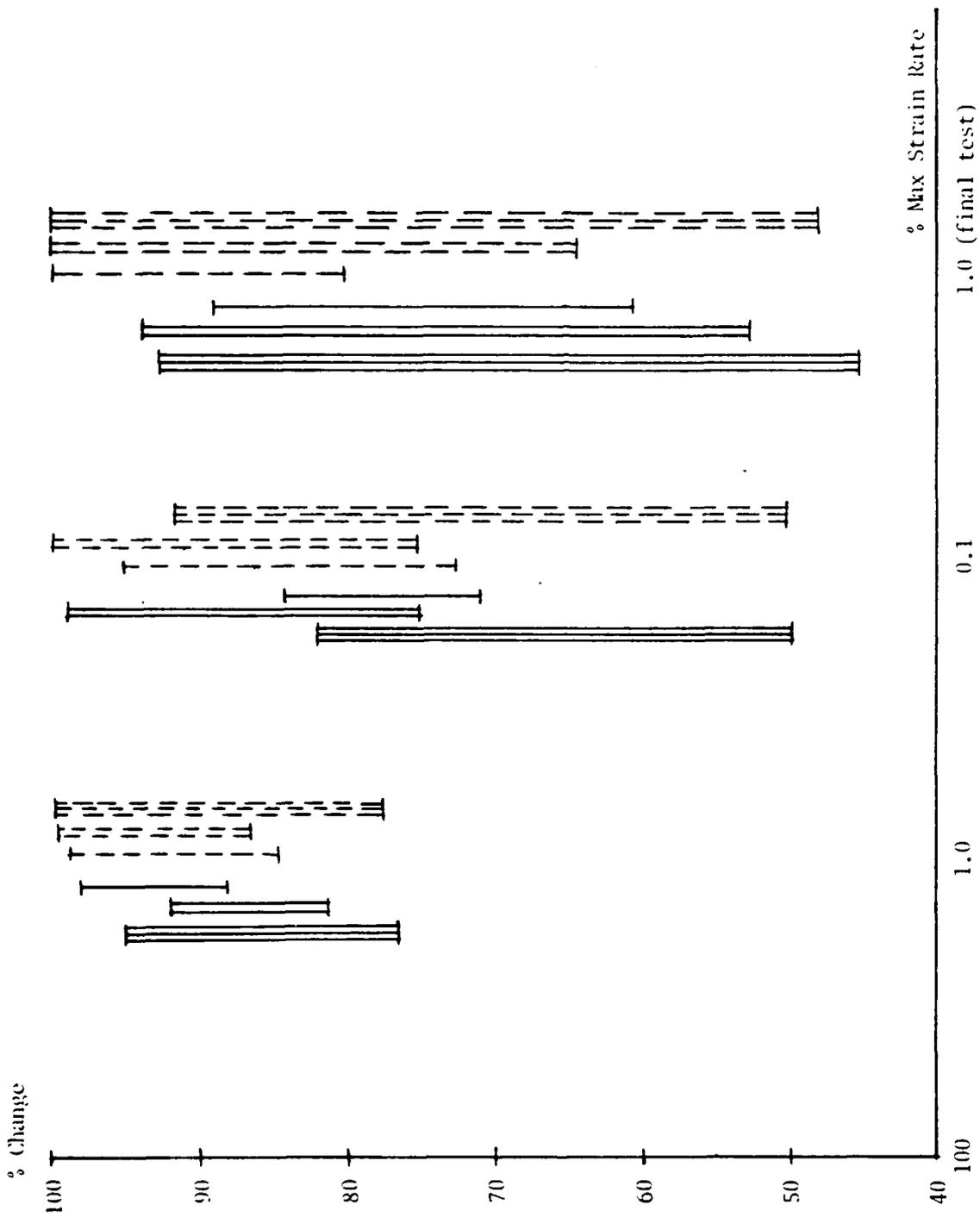


Figure 25: Change in maximum stress and stiffness with strain rate and preconditioning stability for A.I.L.

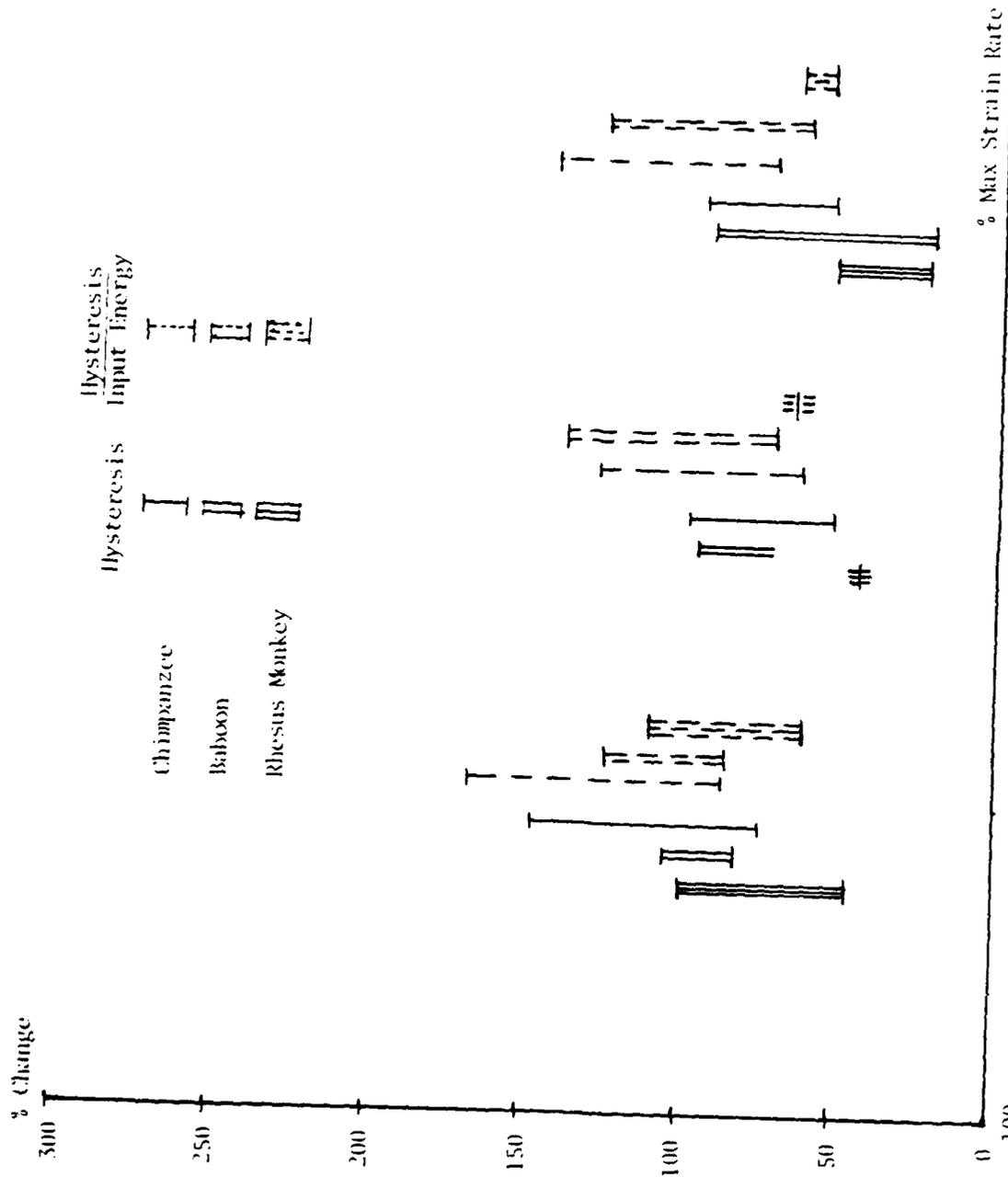


Figure 26: hysteresis dependency upon strain rate and preconditioning stability for S.S.L.

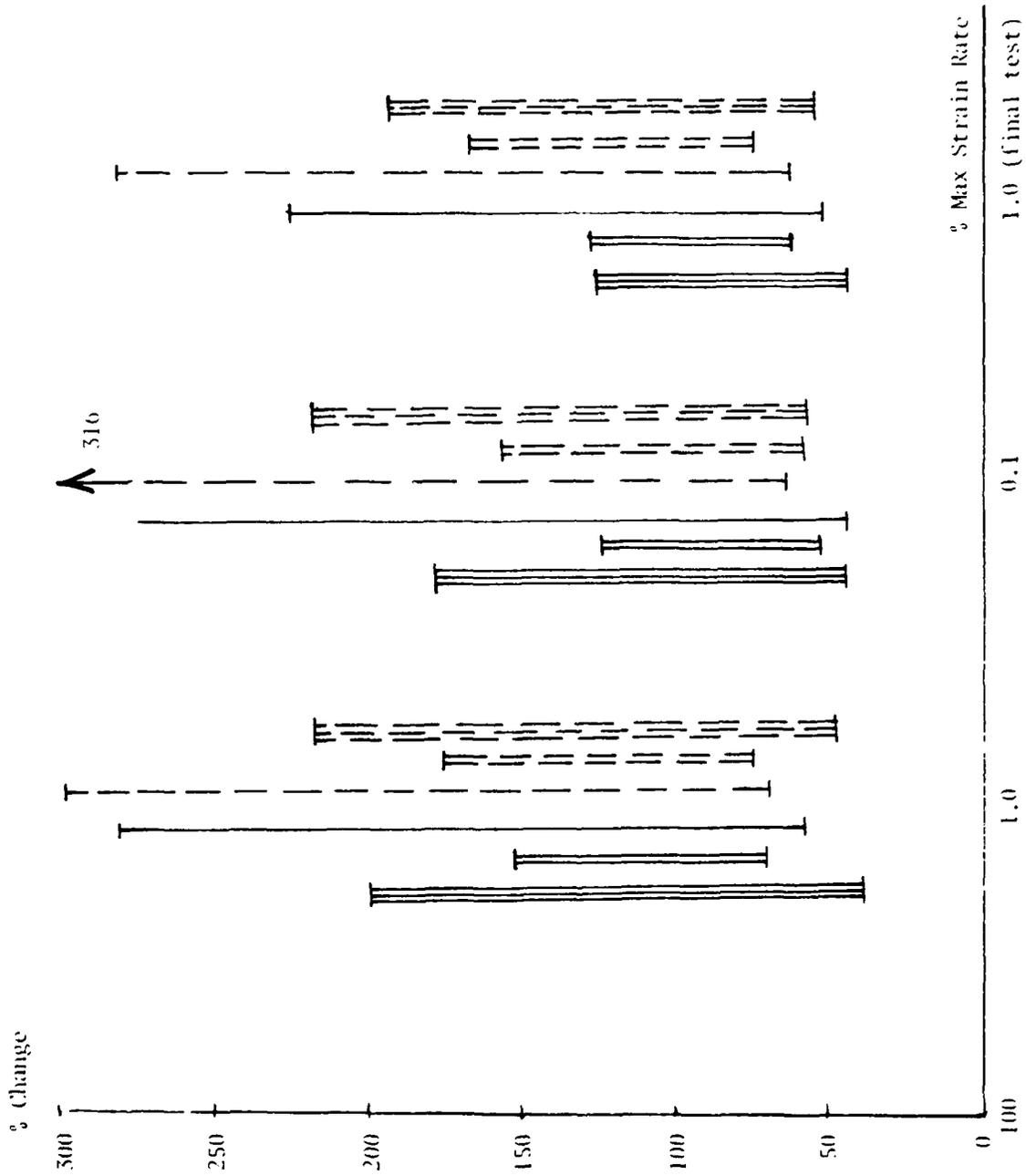


Figure 27: Hysteresis dependency upon strain rate and preconditioning stability for L.F.

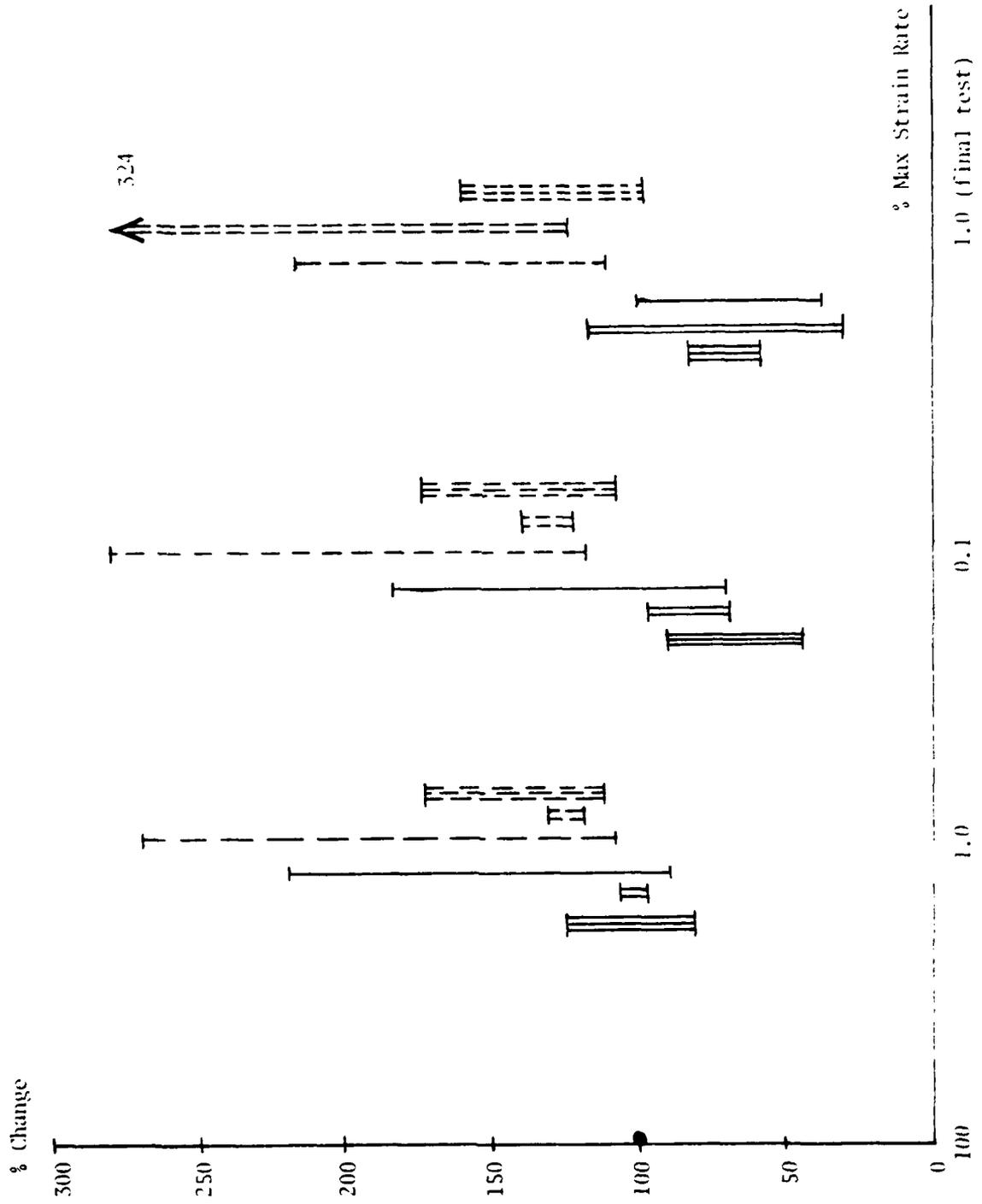


Figure 28: Hysteresis dependency upon strain rate and preconditioning stability for P.L.L.

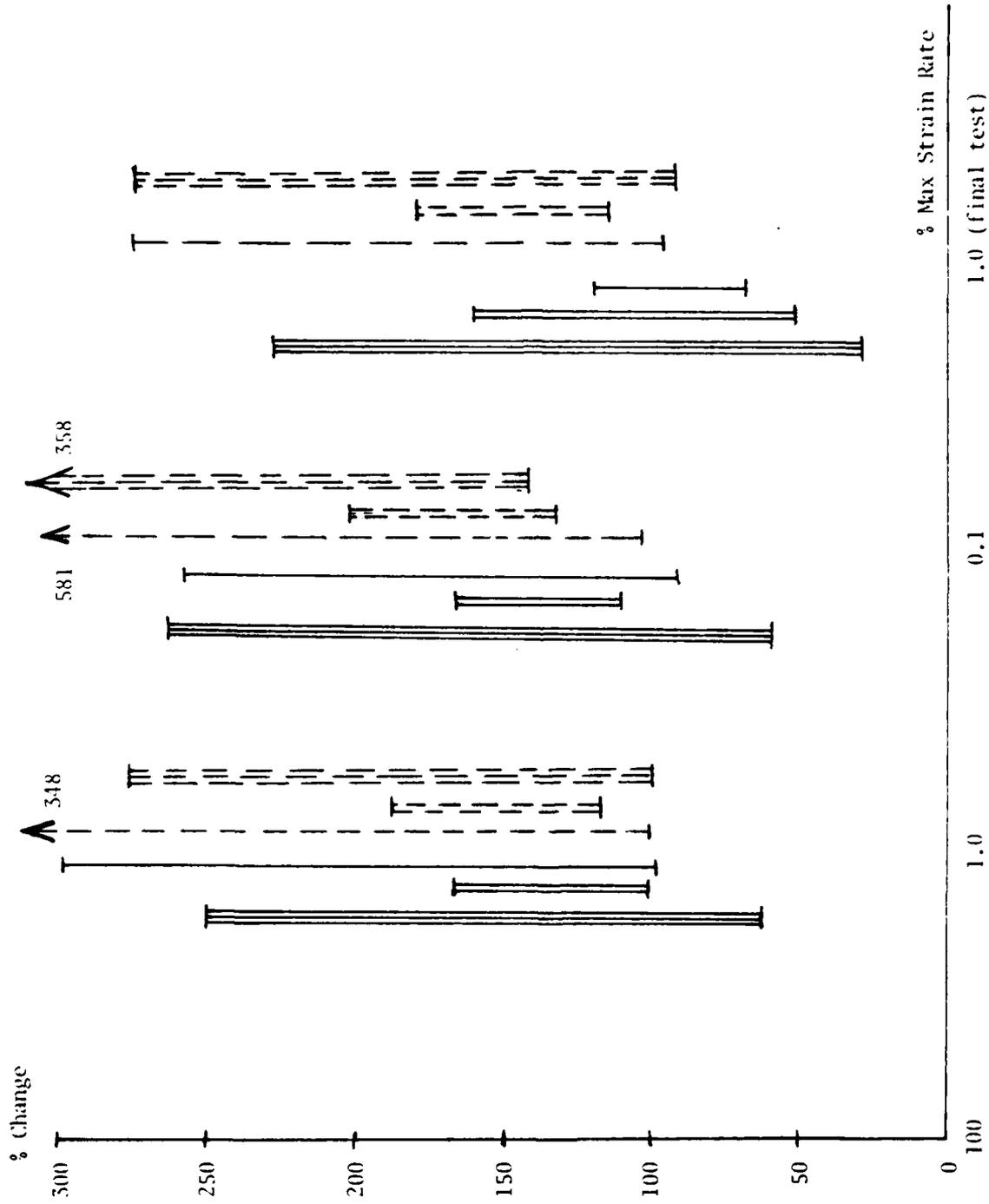


Figure 29: hysteresis dependency upon strain rate and preconditioning stability for A.L.L.

tests and mathematical modeling.

One measurement of prestrain has been recorded involving the intercept of the maximum tangent modulus line with the strain axis. Until additional anatomical measurements are taken, the correlation of this measure with tissue predeformation cannot be made. Additional preconditioning studies and the effects of continued cyclic loading are planned for the next six months. Testing of human spinal ligaments will be initiated as soon as tissue is available.

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