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RHEOLOGY OF FIBRIN CLOTS.
III. Shear creep and creep recovery of fine ligated and coarse unligated clots

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Creep and creep recovery of human fibrin clots in small shearing deformations have been investigated over a time scale from 24 to 10^4 s. Coarse, unligated clots and fine clots ligated by fibrinoligase in the presence of calcium ions were studied to suppliement previous data on coarse ligated and fine unligated clots. Stress was found to be proportional to strain up to at least a maximum shear strain (in torsion geometry) of 2.6%. The initial modulus (25 s after imposition of stress) is proportional to approximately the 1.5 power of concentration for fine ligated and coarse unligated clots. For fine unligated clots, there is comparatively little creep subsequent to the initial deformation; ligation (in this case involving mostly the γ chains) reduces the creep to nearly zero. For coarse unligated clots, there is substantially more creep under constant stress, and creep recovery is not complete. Ligation (in this case involving both γ and α chains) largely suppresses the creep and causes the recovery to be complete. If the structure is fully formed before creep begins, tests of creep recovery by the Boltzmann superposition principle show adherence to linear viscoelastic behavior for all four clot types. Otherwise, the Boltzmann test fails and the recovery is much less than calculated. For fine ligated clots, the observed recovery agrees well with that calculated on the basis of a dual structure model in which an additional independent structure is built up in the deformed state, so that the state of ease after removal of stress is a balance between two structures deformed in opposite senses. It is postulated that the coherence and elastic modulus of the fine ligated clot are largely due to steric blocking of long protofibrils with a high flexural stiffness. In the coarse clot, it is proposed that the structure involves extensive branching of thick bundles of protofibrils, which become permanently secured by the ligation of the α chains of the fibrin.

1. Introduction

Previous papers of this series [1,2] have described measurements of viscoelastic properties of fibrin clots in small deformations. Current related work in other laboratories [3–6] has been largely devoted to measurements of the elastic modulus corresponding to a viscoelastic time scale of a few seconds or minutes, and its relation to fibrin concentration, the kinetics of clot formation, and the role of α and γ ligation by fibrinoligase.

In the preceding paper [2], we noted that studies in the viscoelastic time scale of the order of an hour can provide information about structural rearrangements in fibrin clots. Most of the data reported there concerned coarse ligated and fine unligated clots. Here "coarse" and "fine" refer to clots of high and low opacity, respectively, in which there is much or very little lateral aggregation of the protofibrils which are believed to be the initial product of polymerization of fibrin monomer in a staggered overlapping arrangement. The present paper describes measurements on the other two categories of clots: fine ligated and coarse unligated. The dependence on strain and on elapsed time since clotting is examined. Creep recovery is followed under conditions where mechanical deforma-

tion is imposed either before or after the clot structure has been completely developed.

2. Materials

The sources and methods of processing of purified fibrinogen (human), thrombin (bovine), and fibrin stabilizing factor (FSF, human) were largely the same as in previous papers of this series. For some experiments, the Kabi fibrinogen was further purified by precipitation with β-alanine in a modification of the method described by Straughn and Wagner [7]; the clottability was increased to 95 to 98%. One lot of fibrinogen had been purified by the method of Blombäck and Blombäck [8]. Aliquots of solution were frozen for storage but were not kept for more than one month. In most of the experiments, plasmin was inhibited by the presence of 2 units/ml of Trasylol (FBA Pharmaceuticals, New York). Solutions to be clotted without ligation contained 0.001 M ethylene diamine tetraacetate (EDTA); solutions to be clotted with ligation contained 0.0032 M calcium chloride plus 24 mg/ℓ of FSF (preactivated by thrombin [1]). Presence or absence of ligation was tested by polyacrylamide gel electrophoresis after reduction and solubilization by sodium dodecyl sulfate [9], and sometimes also by solubility in alkaline 40% urea solution. The fine clots were prepared at pH 8.5, ionic strength (μ) 0.45; coarse clots at pH 7.5, ionic strength 0.15. Opacities were measured on separate aliquots as described previously [2]. For the coarse clots they ranged from 2.8 to 6.1 cm^{-1}; for the fine clots, they were never greater than 0.04 cm^{-1}.

3. Method

The use of the Plazek torsion apparatus [10] for measurement of creep and creep recovery of a disc-shaped sample between parallel plates has already been described [2]. Manual rather than automatic recording of the angular deflection was used in most of the experiments reported here. All measurements were made at room temperature, which was between 22 and 23°C.

4. Results

4.1. Dependence of initial modulus on strain

After imposition of stress, the first reading of strain was usually made at about 25 s. The ratio of stress to strain at this moment is designated as an initial modulus; although the subsequent progress of creep is conventionally calculated as compliance (strain/stress ratio), the modulus is the direct measure of clot rigidity. In a series of measurements on a fine ligated clot 26 h after clotting (a sufficient interval to ensure complete development of structure — see section 4.2 below), the dependence of initial modulus on strain was studied as follows. A stress was imposed, a strain reading was taken, the stress was immediately removed, and complete recovery was observed. Repetition for various stresses provided the data shown in fig. 1. Here the word "maximum" applied in the figure to both stress (σ) and strain (γ) refers to the values at the periphery of the disc-shaped sample where they are largest, as calculated by the formulas [11]

$$\sigma_{max} = 2\mathcal{S}/\pi R^3 , \qquad (1)$$

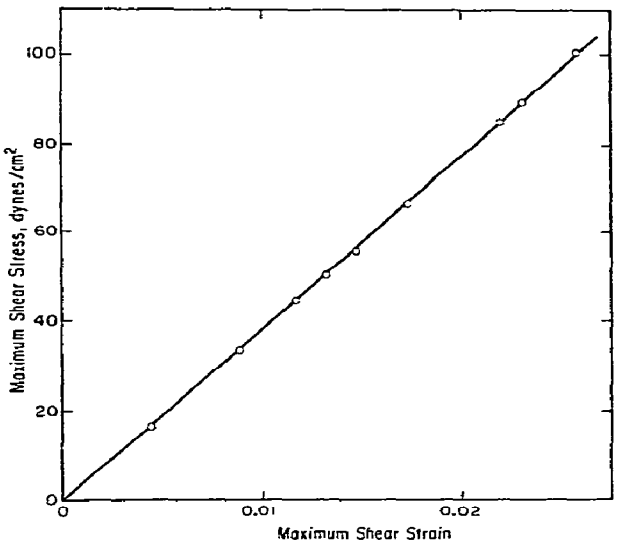

Fig. 1. Plot of shear stress against initial (25 s) shear strain for a fine ligated clot; fibrin 8.0 g/ℓ, pH 8.5, μ 0.45, FSF 25 mg/ℓ, Ca^{++} 0.0032 M, clotting time 11.3 min, age 26 h.

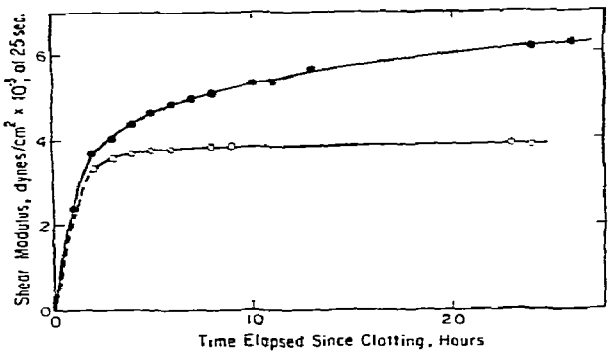

Fig. 2. Plots of initial (25 s) shear modulus against time elapsed since clotting. Open circles, fine ligated, same conditions as fig. 1. Black circles, coarse unligated; fibrin 9.3 g/ℓ, pH 7.0, μ 0.15, EDTA 0.001 M, clotting time 4.8 min.

$$\gamma_{max} = \theta R/h \,, \qquad (2)$$

where $\mathcal{S}$ is the torque, θ the angle of rotation of the upper plate of the apparatus, R the sample radius, and h the sample thickness. The stress and the strain are directly proportional up to a strain of 2.6%, so the modulus (which is $\sigma_{max}/\gamma_{max}$) is independent of strain. Similar linearity not only of initial elasticity but also of viscoelasticity in creep and creep recovery was previously found for fine unligated and coarse ligated clots [2].

4.2. Dependence of initial modulus on time elapsed since clotting

The development of the modulus with time elapsed after the moment of clotting was followed by a sequence of 25-s stress readings, the stress being removed immediately after each measurement with observation of complete recovery. The results for a fine ligated and a coarse unligated clot are shown in fig. 2. The former approaches its limiting value to within 3% after 6 hours (30 times the clotting time); the latter is still increasing somewhat after 24 hours (300 times the clotting time). Gel electrophoresis showed, for the fine ligated clot, γ–γ dimer formation complete in 2 hours but no detectable α ligation even at the end of the experiment; for the coarse unligated clot, there was no α ligation and not more than about 5% γ–γ dimerization even at the end of the experiment.

4.3. Characteristics of creep and effect of ligation

Typical creep curves for coarse and fine clots, ligated and unligated, are plotted with logarithmic scales in fig. 3. Here the creep compliance $J(t)$ is the ratio of time-dependent strain, $\gamma(t)$, to constant stress, σ. For the ligated clots, examples are included at both early and late stages of structural development (cf. fig. 2). Relevant data are listed in table 1. A similar but incomplete comparison in the preceding paper [2] showed the effect of ligation on coarse clots.

For the unligated fine clot, a small amount of progressive creep follows the immediate response to stress, but this is largely suppressed by ligation, even when the structure is not quite fully developed as indicated by the magnitude of the modulus. The ligation involves almost solely the γ chains as indicated by gel electrophoresis. For the coarse clots, as previously noted, absence of ligation is associated with substantial creep; ligation largely suppresses the creep and reduces the magnitude of the compliance (raises the modulus); just as for the fine clots, the modulus increases more when the fully ligated structure is developed. For coarse clots, however, work of Shen and Hermans [4] and McIntire [5] indicates that the modulus increase is associated largely with α chain ligation.

The modulus of a coarse clot is several times as large as that of the corresponding fine clot, whether ligated or not, as reported previously [1,2].

4.4. Dependence of modulus on fibrin concentration

The reciprocal of the compliance at 24 s after imposition of stress is taken arbitrarily as the initial shear modulus, $G_\delta = 1/J(t)$ for $t = 24$. This quantity was determined for various clots with different fibrin concentrations, in three categories: coarse, unligated; fine, partially γ ligated (lack of complete ligation attributed to lower activity of fibrinoligase in this series); and fine, fully γ ligated by additional fibrinoligase plus calcium. The data are listed in table 2 and plotted logarithmically in fig. 4, together with lines reproduced from refs. [1] amd [2].

The modulus is approximately proportional to the 1.5 power of concentration for fine ligated and coarse unligated clots. The new data for the latter agree with those reported in ref. [1], not with the more fragmen-

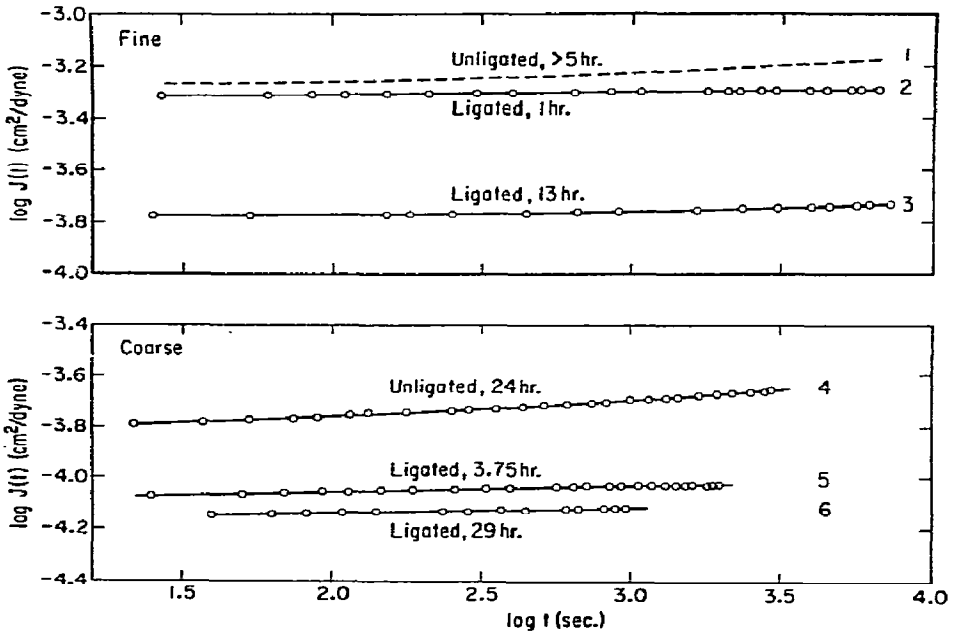

Fig. 3. Creep curves for different types of clots, including effects of ligation at different elapsed times since clotting as shown; conditions are given in table 1.

Table 1
Data for comparison of types of clots (fig. 3)

Curve no.	Expt. no.	Fibrin conc. (g/ℓ)	pH	μ	Clotting time (min)	Time elapsed [a] (h)	Ligation	$-\log J(t)$ initial [b] (dyn/cm^2)	$\frac{d \log J(t)}{d \log t}$ [c]
Fine									
1	d)	(8.1)	8.5	0.45	(5.4)	>5	No f)	(3.27)	0.032
2	94	8.0	8.5	0.45	9.0	1.0	γ g)	3.31	0.01
3	91	8.2	8.5	0.45	10.5	13	γ g)	3.78	0.01
Coarse									
4	82	9.4	7.0	0.15	4.7	24	No h)	3.79	0.065
5	25 e)	8.7	7.4	0.15	7.5	3.75	Yes i)	4.07	0.025
6	25 e)	8.7	7.4	0.15	7.5	29.5	Yes i)	4.15	0.02

a) From moment of clotting to imposition of stress.
b) At 25 s after imposition of stress.
c) At $10^{2.5}$ s after imposition of stress.
d) Interpolated from data of fig. 3, ref. [2].
e) From data of fig. 7, ref. [2].
f) Gel electrophoresis on similar clots showed absence of ligation.
g) Gel electrophoresis showed complete γ chain ligation and trace of α ligation.
h) Gel electrophoresis showed <5% γ ligation.
i) Clots insoluble; presumably both γ and α ligation.

Table 2
Initial moduli of various clots

Expt. no.	Fibrin conc. (g/ℓ)	Clotting time (min)	γ-chain ligation %b)	$-\log J(t) = \log G_\delta$ 24 s (dyn/cm^2)
Coarse, unligated, age 24 h, pH 7.50, μ 0.15, 1 mM EDTA				
65	5.00	6.4	0	3.42
64	6.24	6.9	0	3.58
67	7.59	8.8	0	3.69
63	7.78	8.3	0	3.77
76	7.89	20.2	c)	3.73
78	8.02	20.0	c)	3.79
77	8.13	20.2	c)	3.78
62	8.71	9.3	0	3.80
61	8.86	9.6	0	3.80
84 a)	9.34	4.8	0	3.79
82 a)	9.42	4.7	0	3.79
83 a)	9.58	4.7	0	3.85
86 a)	9.58	4.3	0	3.79
Fine, partially ligated, age 3 to 20 h, pH 8.50, μ 0.45, Ca^{++} 3.2 mM, FSF 30 mg/ℓ d)				
42	3.52	6.5	20	2.46
48	3.58	6.8	40	2.42
50	4.50	6.4	25	2.81
44	5.11	7.3	30	2.78
49	5.30	9.0	30	2.97
41	6.70	23.5	40	3.16
51	7.41	9.6	20	3.34
45	7.64	7.7	25	3.15
47	8.92	7.5	20	3.51
43	10.4	6.7	40	3.55
Fine, ligated, age 10 h, pH 8.5, μ 0.45, Ca^{++} 3.2 mM, FSF 24 mg/ℓ e)				
111	3.27	5.5	100	2.82
108	4.12	13.0	100	2.92
109	5.34	14.0	100	3.17
110	6.65	15.0	100	3.29
113	7.60	15.0	100	3.38
112	9.50	14.0	100	3.57

a) Imidazole buffer pH 7.00, μ = 0.15.
b) Determined by SDS gel electrophoresis.
c) Not measured.
d) FSF not activated by thrombin prior to clotting.
e) This series was made with fibrinogen purified by Dr. B. Blombäck [8].

tary data in ref. [2] which probably do not represent fully developed structures in the light of fig. 2. The data for partially γ ligated fine clots are scattered but generally lie below those for fully ligated. The γ chain ligation was accompanied by not more than a trace of α ligation. Some data for fine unligated clots were reported earlier [2], but they are close to the partially ligated results shown here and it is not certain that ligation was completely absent. Further work on fine unligated clots will be reported subsequently.

Some fibrinogen preparations without addition of Trasylol gave clots with abnormally low moduli, attributed to some damage by digestion by contaminant plasmin [12]. One preparation gave abnormally *large*

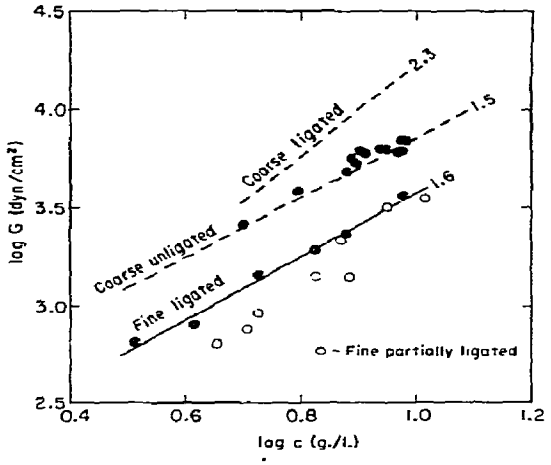

Fig. 4. Initial shear modulus plotted against fibrin concentration. Points, data from table 2; open circles, partially ligated fine clots; dashed lines reproduced from ref. [2]. Numbers at right denote slopes.

moduli especially at low concentrations. These data are not reported. The sensitivity of elastic modulus to small modifications in the fibrinogen has been stressed by Shen and Hermans [4,12].

Some different concentration dependences of modulus have been reported by other authors. Shen and Hermans [4] found it to be proportional to c^2, both ligated and unligated, but in a lower concentration range (<1 g/ℓ) where the clot structure may be qualitatively different. Glover and McIntire [5] reported direct proportionality to c, but they worked with plasma fortified by dry fibrinogen lyophilized from a salt solution, so large amounts of other proteins were present and the ionic strength was changing together with the fibrinogen concentration.

4.5. Course of creep and recovery

After imposition of constant stress, the strain was repeatedly measured during creep over times up to several hours. In some experiments, creep recovery measurements were subsequently made by rotating the top of the torsion wire back to the position corresponding to zero torque and following the decrease in strain with time. Examples of stress and strain plotted against time are shown in figs. 5 and 6 for a fine ligated and a coarse unligated clot respectively. Here the time variable (elapsed since imposition of stress) must be clearly distinguished from that in fig. 2 (elapsed since moment of clotting, with measurements always 25 s after imposition of stress). The terms "maximum stress" and "maximum strain" on the ordinates of figs. 5 and 6 have the same significance as in fig. 1.

Before creep recovery, as shown on the graphs, the

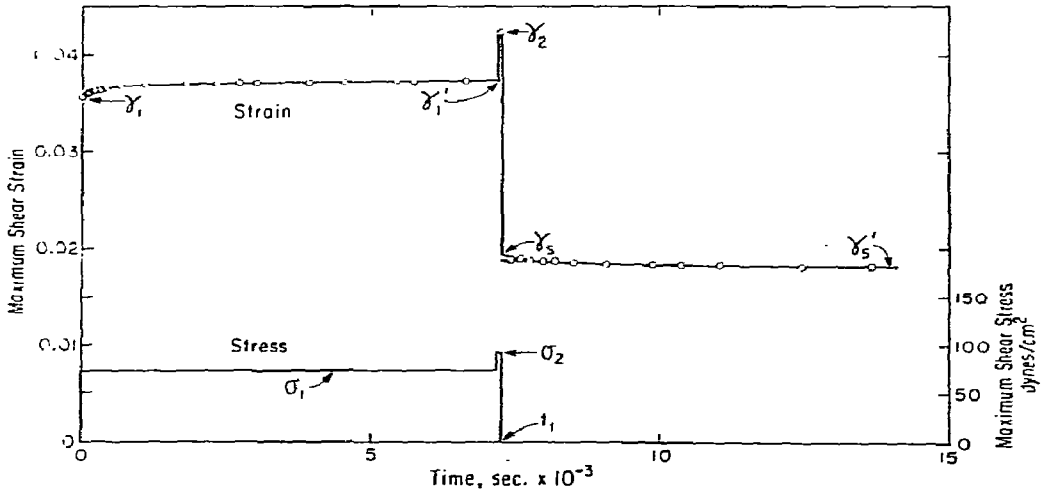

Fig. 5. Time profile for stress and strain in creep and creep recovery experiment, fine ligated clot; fibrin 8.0 g/ℓ, pH 8.54, μ 0.45, FSF 24.6 mg/ℓ, Ca^{++} 0.0032 M, clotting time 9.0 min, opacity 0.02, age at start of creep 1.0 h.

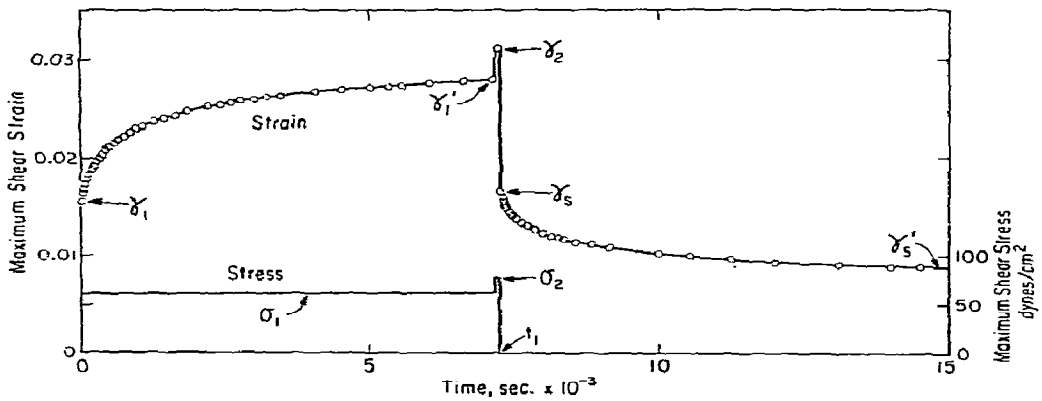

Fig. 6. Time profile for stress and strain in creep and creep recovery experiment, coarse unligated clot; fibrin 9.6 g/ℓ, pH 6.96, μ 0.15, EDTA 0.001 M, clotting time 4.3 min, opacity 5.9, age at start of creep 2.0 h.

stress was increased from σ_1 to σ_2 for about 25 s and the corresponding increase in strain from γ_1' to γ_2 was measured. The purpose of these additional measurements is explained in the Discussion. The failure to attain complete recovery, seen in both figs. 5 and 6, is characteristic of clots subjected to shear creep at a relatively early age (i.e., elapsed time since clotting), 1 h and 2 h, respectively. When 12 to 24 hours were allowed to elapse since clotting before imposition of shear stress, the subsequent creep recovery was greater, and for fine ligated clots it was nearly complete as will be shown below.

In analyzing the behavior shown in figs. 5 and 6, we distinguish the levels of stress and strain indicated on the graphs by arrows. Here γ_1 and γ_1' are the strains measured initially and finally under stress σ_1; γ_2 is the initial strain under σ_2; γ_s and γ_s' are the initial and final strains measured in recovery after removal of stress. These quantities are somewhat arbitrary, depending on the time pattern of the experiment, especially for the coarse unligated clot of fig. 6. For the fine ligated clot, both deformation and recovery are rapid, with very little delayed creep; for example, $\gamma_1/\gamma_1' = 0.95$. For the coarse unligated clot, the time-dependent strain is much greater; $\gamma_1/\gamma_1' = 0.55$. The latter behavior agrees with that previously reported for coarse unligated clots [2], plotted differently with doubly logarithmic scales; for coarse ligated clots, both creep and creep recovery are largely accomplished in brief time intervals following changes in stress. For fine clots, ligation makes less difference in the course of creep and creep recovery, as seen in fig. 2. Several other creep and recovery experiments were analyzed in a similar manner and compared with calculations of recovery from a dual structure model as described in section 5.3 below.

4.6. Tests of Boltzmann superposition principle for clots with modulus fully developed before stressing

If there are no structural changes associated with either strain or aging and the viscoelastic behavior is linear, the Boltzmann superposition principle should permit calculation of not only the maximum recovery (γ_s') but also the entire course of the recovery curve, $J_r(t)$, from a duplicate experiment in which creep is measured over a time span encompassing that for which recovery is to be predicted:

$$J_r(t) = J(t) - J(t - t_1)\,, \tag{3}$$

where t_1 is the time at which stress was removed and $J(t)$ is the creep measured in the duplicate experiment in which stress is not removed.

This test was previously found to be successful [2] for coarse, ligated and fine, unligated clots provided the clot age at the beginning of creep was sufficient for the structure to have been fully developed. It is illustrated for a fine ligated and a coarse unligated clot in figs. 7 and 8 respectively. Here, unlike figs. 5 and 6, the ordinate is logarithmic. Also, the clots are aged to permit full development of structure. It may

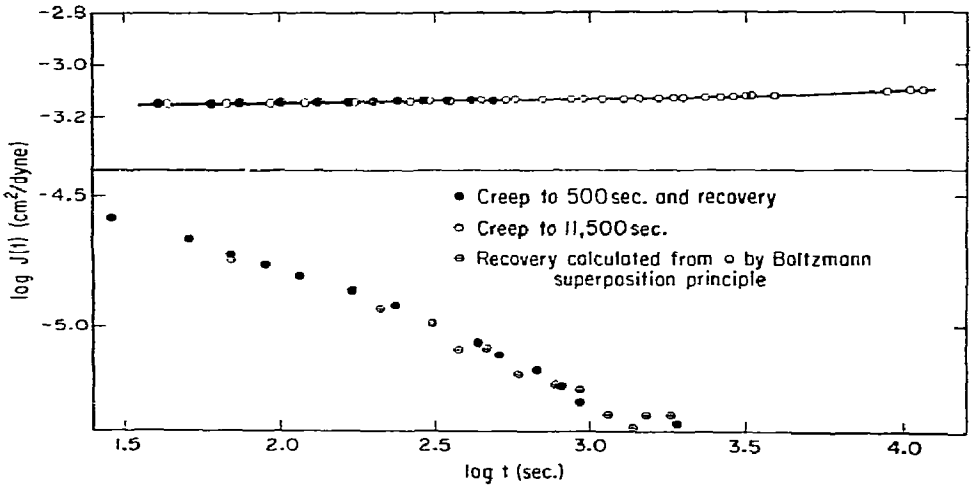

Fig. 7. Creep compliance and creep recovery compliance for a fine ligated clot with test of Boltzmann superposition principle. Fibrin 6.7 g/ℓ, pH 8.5, μ 0.45, FSF 29.7 mg/ℓ, Ca^{++} 0.0032 M, clotting time 23.5 min, opacity 0.04, age at start of creep ca. 6 h, γ chain ligation 40%.

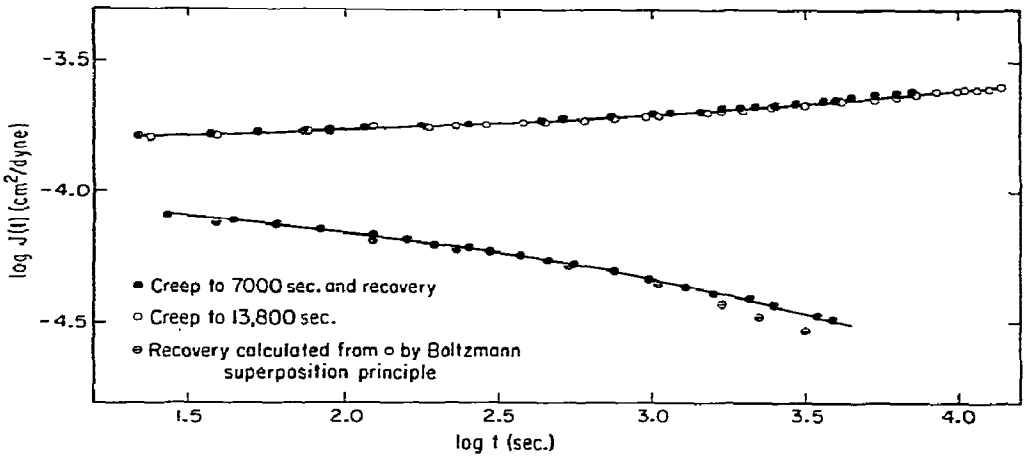

Fig. 8. Creep compliance and creep recovery compliance for a coarse unligated clot with test of Boltzmann superposition principle. Fibrin 9.5 g/ℓ, pH 7.0, μ 0.15, clotting time 4.5 min, opacity 6.1, age at start of creep 24 h.

be noted first that the recovery is more than 99% complete for the fine ligated clot (the residual strain at the end of the recovery experiment being 0.008 of that at the end of the creep experiment); similar results were obtained for fine unligated and coarse ligated [2]. For the coarse unligated clot, recovery is about 88% complete. The course of recovery calculated from equation (3) agrees within experimental accuracy throughout with that observed, for both types of clots.

5. Discussion

5.1. Creep recovery in clots strained before structure is complete; dual structure model

In contrast to the nearly complete recovery seen in figs. 7 and 8 for clots whose structure was essentially completely formed before straining, there is substantial residual deformation for clots subjected to strain at an early age, as seen by the level of γ'_s in figs. 5 and 6.

For such clots, the Boltzmann superposition principle is very far from describing the creep recovery, as already noted previously with coarse ligated clots strained before completion of structure formation [2].

A possible interpretation of this behavior is that additional structure is built up after imposition of stress; this latter structure is isotropic in the deformed state and tends to maintain the deformed shape. Upon release of stress, the clot comes to a state of ease γ_s' in which the two structures pull in opposite directions and balance.

The theory for the dual structure model has been worked out for rubbers with two stages of cross-linking [13,14] and has been applied to stress relaxation studies of gelatin gels [15] and rubbers cross-linked in strained states [16]. It can be formulated for our creep experiments without making any assumptions about the molecular mechanism of elasticity, as follows. In this oversimplified description, we ignore the fact that the deformation is actually changing somewhat during the creep phase, i.e., the new structure is built up at strains which range from γ_1 to γ_1'. For fine ligated clots, this is not serious since the difference between γ_1 and γ_1' is slight (fig. 5). For coarse unligated clots, it causes the predicted recovery to be too small by the order of a few per cent.

Referring to figs. 5 and 6, the shear modulus of the original structure when the creep experiment begins is $G_1 = \sigma_1/\gamma_1$. At the end of the creep period, the brief additional stressing measures the sum of the moduli of the two structures: $(\sigma_2 - \sigma_1)/(\gamma_2 - \gamma_1') = G_1 + G_2$. Thus the modulus of the second structure is

$$G_2 = (\sigma_2 - \sigma_1)/(\gamma_2 - \gamma_1') - \sigma_1/\gamma_1 . \quad (4)$$

In the state of ease, the stresses of the two structures balance:

$$G_1\gamma_s' = G_2(\gamma_1' - \gamma_s') . \quad (5)$$

Hence the fractional recovery is

$$(\gamma_1' - \gamma_s')/\gamma_1' = G_1/(G_1 + G_2) . \quad (6)$$

According to the Boltzmann superposition principle, which should hold if no additional structure is formed during creep, the fractional recovery can be calculated from eq. (3) with $\gamma_1' = \sigma_1 J(t_1)$ and $\gamma_s' = \sigma_1 J_r(t_2)$ where t_2 is the total time elapsed during the experiment.

In table 3, observed recoveries are compared with those calculated by eq. (6) for fine ligated and coarse unligated clots with various ages at the start of creep, together with a few Boltzmann calculations. The latter do not agree with observed values when the creep begins at an early age, as expected.

For the fine ligated clots, the dual structure model agrees very well with the observed recovery. It may be

Table 3
Creep recovery in clots strained before structure is complete

Expt. no.	Age at creep start, (h)	γ_1/γ_1'	G_2/G_1	Fractional recovery, $(\gamma_1' - \gamma_s')/\gamma_1'$		
				Observed	Dual model (eq. (6))	Boltzmann (eq. (3))
Fine ligated						
95	0.75	0.93	3.6	0.22	0.22	
96	1	0.96	1.14	0.50	0.47	
94	1	0.95	0.91	0.51	0.52	
89	1.5	0.93	0.186	0.79	0.84	0.96
97	2	0.92	0.266	0.81	0.79	
91	13	0.90	0.040	0.99	0.96	
Coarse unligated						
85	2	0.55	0.37	0.68	0.73	
80	3	0.56	0.115	0.76	0.90	0.91
76	24	0.67	0.118	0.87	0.89	
82	24	0.67	0.136	0.88	0.88	0.92

concluded that the additional structure built up in the strained state acts independently of the original structure with opposing stresses which balance in the state of ease. For the coarse unligated clots, the calculated recovery is larger than the observed, as though the additional structure were less effective in opposing the original structure than indicated by the increase in modulus from G_1 to $G_1 + G_2$. If a correction is made for the formation of the additional structure during varying strain, the discrepancy is increased slightly.

5.2. Comments on fine clot structure

The ligated fine clot with completed structure exhibits relatively little creep and almost complete recovery. If its fibrillar structure consists of the protofibrils with twice the cross-section area of the monomer and the staggered overlapping arrangement proposed for intermediate fibrin polymers [17], there is no obvious mechanism for branching to form a three-dimensional network structure. However, branching may not be necessary if each fibril extends for a great length, crossing many other fibrils at random angles, and possesses substantial flexural stiffness. A macroscopic analog would be a random assembly of long stiff wires. The shear modulus of the assembly would be proportional to the flexural stiffness of a single wire and some power of the concentration (volume fraction occupied by the wires) dependent on the statistics of the steric interference of the crossing wires. If the flexural stiffness is high, the assembly should be quite coherent even in the absence of cohesive forces at the crossing points, though such forces might be present. Measurements of dilute solution viscoelasticity of intermediate fibrin polymers [18] indicate that their flexural stiffness is quite high, and a striking feature of electron micrographs is the straightness of thin fibrils over great lengths.

It may be remarked that such an assembly of randomly oriented crossing thin rods should be thermodynamically unstable, as shown by calculations of Flory [19], tending to undergo phase separation into a dilute isotropic and a concentrated ordered phase. However, once the structure is established by the rapid linear growth of the polymers in random directions, steric features would make it quite impossible for it to rearrange and separate into two phases. As a matter of fact, if the structure if broken up afterwards by sonication, the fragments do aggregate [20]. There appears to be little if any α ligation in fine clots, though if present it could provide cross-links between fibrils in contact which would strengthen the purely steric coherence of the structure.

In the state of ease of fine clots described in table 3, exemplified in fig. 5, we picture two interpenetrating arrays of such fibrils, one established by linear growth in the original isotropic state, the other by growth in the state of strain during the creep experiment, and both deformed but in opposite senses in the state of ease so that their stresses balance.

5.3. Comments on coarse clot structure

The coarse clot has much thicker fibers and therefore far fewer of them. In the light of the wire structure analog, there is less steric blocking and the structural coherence is presumably established by branching instead. Such branching is seen in electron micrographs and is understandable as the divergence of lateral aggregates. In the absence of ligation, the lateral forces are somewhat weak and structural rearrangements are possible, so that there is substantial creep under applied stress and recovery is incomplete even when the original structure is essentially fully established before creep begins. With ligation (and it is presumably α ligation that is important here, though this remains to be established), the lateral junctions between protofibrils are fixed, creep is largely suppressed, and recovery becomes complete. At the same time, the modulus is increased and its dependence on concentration appears to be modified. At present, these features are not understood because the mechanism of energy storage in the network of thick fibers is in doubt and we are not yet prepared to speculate about it.

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