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THE VISCOUS-ELASTIC PROPERTIES OF THE VITREOUS BODY AND ITS REACTION TO EXTERNAL FORCES

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IN those clinical conditions wherein the behaviour of the vitreous body is of importance—its break-down into a fluid state in inflammatory or degenerative conditions, its swelling in glaucoma, and so on—an accurate measurement of the consistence of its structure is essential if any progress is to be made in the elucidation of the physical changes which determine its conduct. Morever, if it is to be suggested that traction by the vitreous upon a weakened area of the retina may in some cases play a determining part in the aetiology of detachment of the retina, the nature and extent of the force which the vitreous gel is capable of instituting, becomes of considerable interest. It would seem, therefore, that a study of the viscous and elastic properties of the gel is not without importance; and, incidentally, it has proved of interest in so far as it confirms the conclusions published by one of us three years ago (Duke-Elder, 1930) with regard to the intimate physical structure of the vitreous body and its affinity to other gels, such as gelatine.

The very small rigidity of the vitreous body, and the ease with which its structure is broken down by mechanical stresses, make ordinary methods inapplicable to the study of its elastic properties. The work of Lobeck (1929) and Baurmann (1929), in whose experiments external force was applied to the gel, should be interpreted in this sense, for *in vitro* the gel breaks down if subjected to pressures as low as 1 or 2 mm. of water, a phenomenon which occurs if it is merely left unsupported in mass for any length of time. This inability to maintain its physical gel structure when forces are applied to it makes a study of the reaction of the vitreous to forces difficult, but a suitable method was found in a modification of that used by Freundlich and Seifriz (1923-4) in their work on inorganic gels. The essential principle of the method is to introduce a nickel particle into the body of the gel and subject this particle to the force exerted by an electro-magnet; the extent of the excursion of the particle in the field of force, and its recoil when the force is removed gives a measure of the viscosity of the gel, the degree of its consistence, and its elasticity.

The technique finally adopted was as follows:—A small mass of vitreous was introduced with full aseptic precautions into a glass cell made from a piece of glass tubing, 6 mm. long and 10 mm. in diameter, ground square at the ends and cemented to a microscope slide. It could be closed on the top by means of a cover-slip, and made air-tight with vaseline to prevent contamination or evaporation. This was clipped to the stage of a microscope fitted with a Leitz micrometer eye-piece, in which the cross-hair was moved by a screw, bearing a drum divided into 100 divisions. Calibration with a Zeiss object-micrometer showed that, using an objective of 16 mm., one division on the drum was equivalent to $0{\cdot}472 \pm 0{\cdot}001\mu$. The nickel particle was introduced into the mass of vitreous by means of a micromanipulator of the ordinary pattern, made to clamp on to the stage of the microscope; three screws gave movements in three directions at right angles, and several coarse adjustments were also provided. The magnet consisted of an iron core, 54 cm. long and 25 mm. in diameter, on which was wound a coil of No. 24 S.W.G. enamelled copper wire, to a resistance of 280 w. The coil was connected, through a rheostat, ammeter and switch, to the 200 volt D.C. mains. To the end of the magnet core was screwed a horizontal iron rod, 5 cm. long and 6 mm. in diameter.

Some of Kahlbaum's pure nickel powder was placed on a microscope slide and distributed by tapping, the excess being shaken off. The slide was placed on the microscope stage, a particle about 50μ in diameter and as nearly spherical as possible being brought to the centre of the field. The needle in the micromanipulator was then dipped in glycerine and slowly withdrawn and then its point was brought fairly close to the particle selected. Under the microscope the needle was lowered so that its point touched the top of the particle, and then raised, when the particle usually adhered to the point.

In the introduction of the nickel particle into the vitreous, if the needle with the particle were merely immersed in the gel it was found that the particle did not normally come away from the needle in the gel, and, moverover, that the withdrawal of the needle caused a considerable disturbance, often lifting the mass of vitreous bodily out the cell. The micromanipulator was, therefore, employed merely to place the particle on the surface of the gel. The cell was then closed with a cover-glass and placed on the top of the field magnet of a string galvanometer, so that its centre was just over the gap beween the poles. The current was switched on, and after about an hour, the particle was found to have penetrated 1 or 2 mm. below the surface.

After the particle had been introduced it was necessary to wait for an hour or two for the system to attain equilibrium. The cell was then placed on the microscope stage and arranged so that one edge of the particle was seen to coincide with the zero mark of the micrometer eyepiece and measurements were made of the displacements produced on exciting the magnet.

When the end of the magnet was brought near to the cell and the current switched on, the particle could be seen to move, and to spring back towards its original position on switching off, thus showing that the gel possesses real elasticity. On the other hand, bringing the unexcited magnet near the cell produced no perceptible effect, showing that the residual magnetism of the core could be neglected. It was found, however, that, if the current was allowed to flow only for a very short time (*e.g.*, 1/5 second), displacements could be obtained which were completely and instantaneously reversible, but that, if the current were left on for any appreciably longer time, the return of the particle was, in general, incomplete. Both the reversible and the irreversible components of the displacements were found to increase with the time for which the current was allowed to flow, a typical set of results being shown in Table 1.

It is clear from these figures that the vitreous is not behaving like an ordinary elastic body, and it appears that its viscous properties must be taken into account. The fact also that displacement No. 6 differs so greatly from No. 1 shows also that the behaviour of the gel is dependent on its previous history.

In order to analyse the motion of the particle in the vitreous humour, arrangements were made for observing the position of the particle at known intervals of time after switching on the magnet. A typical example of the curves so obtained with cat's vitreous is shown in Fig. 1. It will be seen that the curve is initially very steep indicating a rapid acceleration and an almost instantaneous displacement, but that it soon bends over, the final configuration being practically straight, corresponding to a uniform motion of the particle.

TABLE I—CAT'S VITREOUS

	Time for which magnet was excited.	Reversible Component.	Irreversible Component.
		Scale Division.	Scale Division.
1	10 seconds	14·1	0·0
2	30 seconds	14·7	1·0
3	1 minute	17·4	1·6
4	3 minutes	23·1	6·0
5	9 minutes	35·0	16·3
6	10 seconds	26·6	1·4

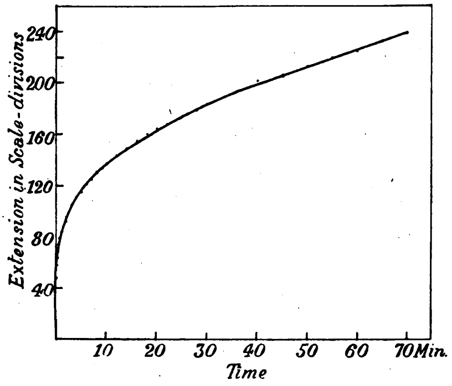

FIG. 1.

Vitreous humour from ox eyes appears to have much the same elastic properties as that obtained from cats, although it is definitely less rigid, and gives curves of a slightly different shape. Fig. 2*a* shows the time extension curve obtained with ox vitreous, the end of the magnet being at a distance of 15 mm. from the particle. Comparison with Fig. 1 (cat's vitreous, 8 mm.) will show that, although in the case of the ox vitreous the field was weaker, the displacement produced was much greater, while the curve in Fig. 2*a* bends over considerably more slowly than that in Fig. 1. The size of nickel particle was approximately the same in both cases, and the current flowing in the magnet was identical, so that the experiments are comparable even if not quantitatively the same. The differences exhibited appear to be inherent ones, as the respective curves are not isolated examples, but typical of many experiments with different specimens.

A consideration of these time-extension curves is of considerable interest, and the two types of extension, the initial rapid movement and the subsequent slow "creep," find their counterpart in the behaviour of other inorganic gels which, like the vitreous, are made up of minute fibrillar micellae suspended in and associated with a viscous fluid (*e.g.*, gelatine, Poole, 1925). The stretching of a gel system of this nature consisting of a network of elastic fibrils suspended in a viscous liquid should be made up of two components—an instantaneous extension due to the sudden stretching of the fibrils, followed by a slower creep, during which the intermicellar liquid readjusts its position by viscous flow.

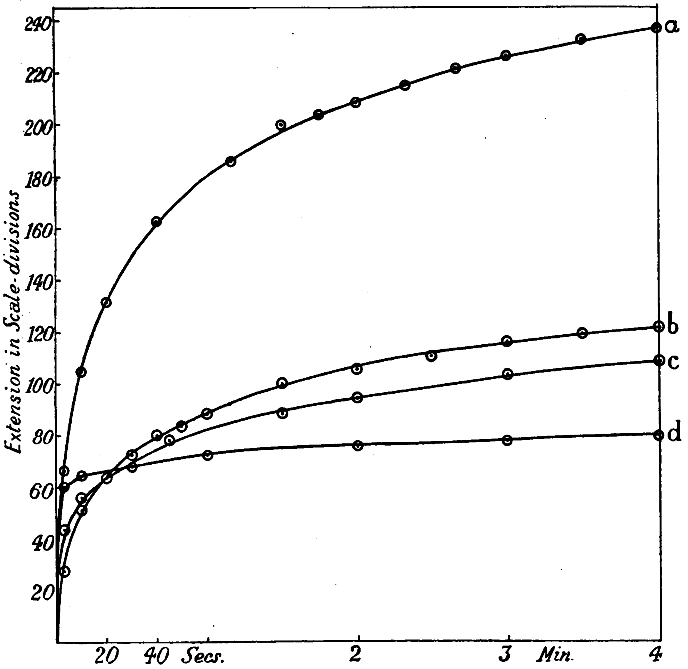

FIG. 2.

If, instead of plotting a simple time-extension curve, a derived curve is obtained wherein increments of extension (dE/dT) are plotted against total extension (E), further interesting considerations arise. Such a derived curve from Fig. 1 is seen in Fig. 3;

it is straight over a considerable portion of its length, but later undergoes a sudden inflection, the remainder being curved, thereafter tending to become asymptotic to the E axis. The general form of these curves is very similar to that of Poole's curves for gelatine, the discrepancy shown by the first point being doubtless due to the fact that the system had not settled down to a steady state after the first instantaneous displacement. This discrepancy of the first one or two points was almost always found to be present.

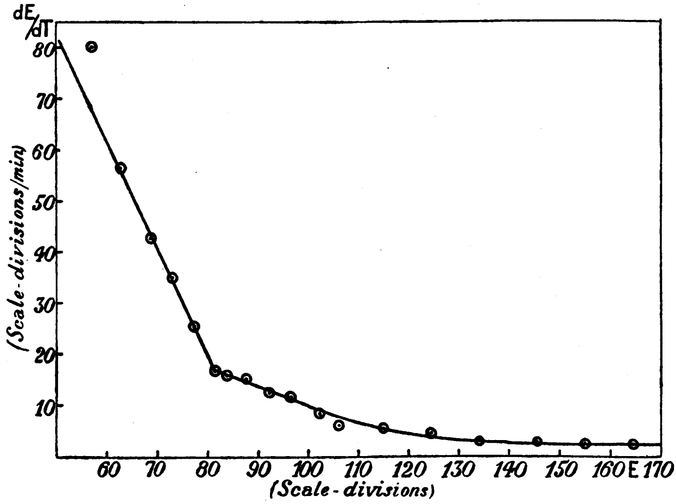

FIG. 3.

Theoretically this "viscous-elastic creep" should be in accordance with the equation

$$K_1E + K_2\eta(dE/dT) = \text{constant} \qquad (1)$$

where

K_1 is a constant proportional to the elasticity of the fibrils.
K_2 is a factor depending on the nature of the channel through which flow can occur.
η is the coefficient of viscosity of the intermicellar liquid.
E is the total extension, and
T is the time taken to produce this extension.

Transposing (1) and differentiating with respect to E,

$$\frac{d(dE/dT)}{dE} = -\frac{K_1}{K_2\eta} \qquad (2)$$

In other words, the curve representing equation (1), with E and dE/dT as variables, should be a straight line, the slope of which is inversely proportional to the viscosity of the liquid and to a factor depending on the nature of the flow-path, while being directly proportional to the modulus of elasticity of the fibrillae. Since neither the dispersion nor the viscosity should be affected by variation of the load, the slope of the curve of equation (1) should be independent of the load. The fact that the slope of the straight portion of the $d\mathrm{E}/d\mathrm{T}$—E curve is independent of the force acting on the particle was shown by carrying out successive experiments with the magnet at different distances from the particle. In Fig. 4 is shown the $d\mathrm{E}/d\mathrm{T}$—E curve obtained with the end of the magnet at a distance of 12 mm. from the nickel particle, and Fig. 5 shows the corresponding curve obtained when this distance was reduced to 8 mm. Neglecting the first two points, the slopes of the straight portions of two curves are found to be the same within experimental error, the value of $d^2\mathrm{E}/d\mathrm{T}^2$ being

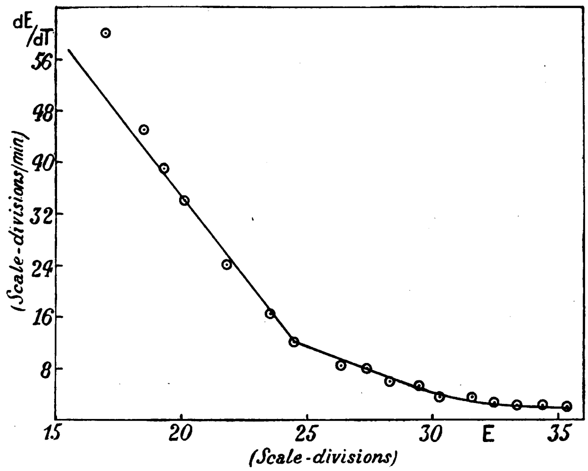

FIG. 4.

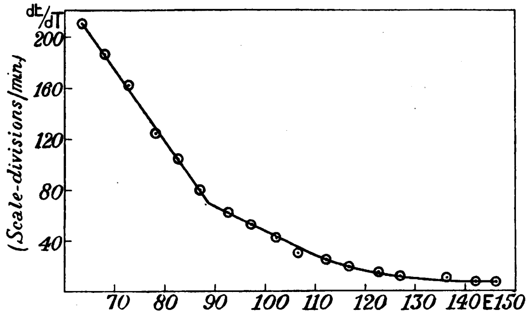

FIG. 5.

5·1 minutes. Fig. 6*a* shows the dE/dT curve derived from Fig. 2*a*, the slope of the straight portion is 6·1 minutes. Figs. 2*b* and 6*b* show the corresponding curves for v = 20 mm.; in this case the straight portion of the derived curve had a slope of 6·8 minutes—the same within experimental error.

Up to this point, therefore, the displacement of the nickel particle in the vitreous appears to follow the same laws as the linear extension of gelatine and constitutes strong evidence in favour of the micellar theory of the structure of the vitreous gel.

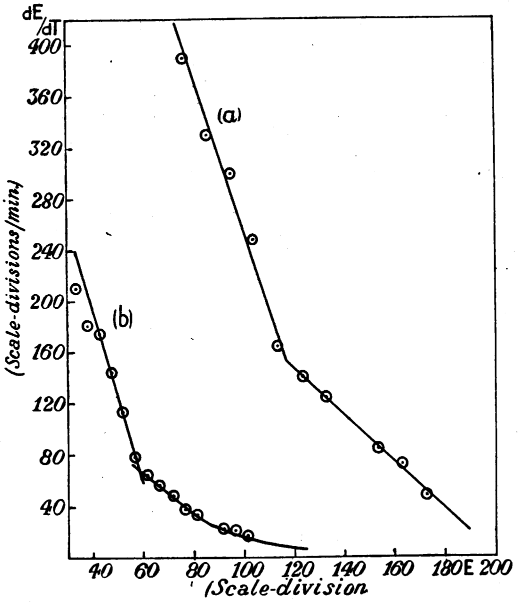

FIG. 6.

Poole also found that the extension corresponding to the point of inflection of the dE/dT—E curve was equal to the greatest reversible extension for the load in question, and was directly proportional to that load. The rate of return of the particle in the vitreous, after switching off the magnet, is so slow that the determination of its final equilibrium position is rather uncertain without careful control of temperature, etc. In the two cases, illustrated in Figs. 4 and 5, the reversible components of the displacements appeared to be 38·2 and 100·8 scale divisions respectively, while the extensions corresponding to the points of inflection of the dE/dT—E curves are considerably smaller, *viz.*, 24·5 and 88·8 scale divisions.

Acidification of the vitreous produces a considerable change in the shape of the T—E curve. Fig. 2*c* shows a curve obtained with ordinary ox vitreous, while in Fig. 2*d* is shown the curve obtained after addition of a drop of very dilute hydrochloric acid, the other conditions remaining the same. It will be seen that the instantaneous displacement is much increased, while the "creep" is greatly diminished, so that the gel behaves more like an ordinary elastic solid. The reason for this effect is doubtless the shrinkage of the gel and withdrawal of water from the fibrillae, although the acidification was not sufficient to modify appreciably the ultra-microscopic appearance of the gel.

In a previous paper (1932) it was shown that dilute plasma clots had an appearance, an ultramicroscopic structure, and physical

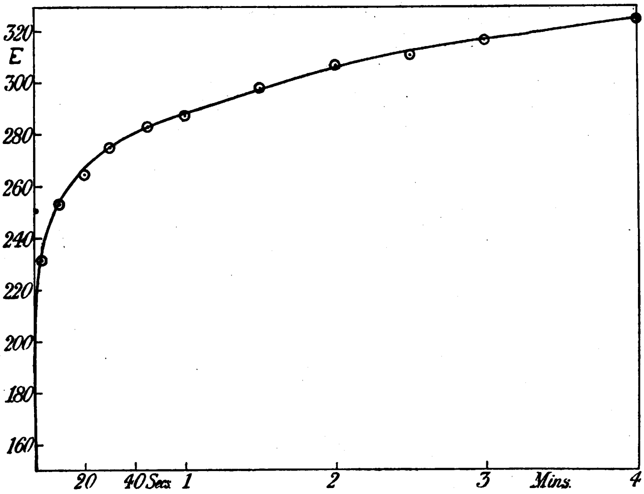

FIG. 7.

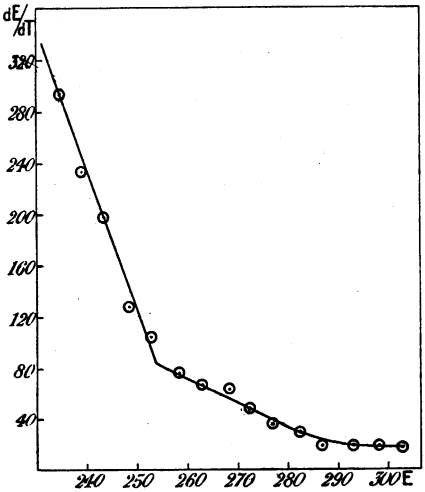

FIG. 8.

properties very similar to those of the vitreous gel. It is interesting that the analogy is further confirmed by its general elastic behaviour. When plasma clots were used, on starting the current in the magnet, the particle suffered an instantaneous displacement, followed by a slow viscous-elastic creep. The instantaneous component of the displacement was, however, a much greater fraction of the whole than in the case of the vitreous body, and the rate of creep diminished much more rapidly. This considerably increased the difficulty of observing the initial phases of the displacement.

The time-displacement curves were similar to those obtained with the vitreous body, but the $d\mathrm{E}/d\mathrm{T}$—E curves were somewhat inconsistent. As a rule they were of the usual type—a straight initial portion, followed by a well-defined break, the curve then becoming asymptotic to the E axis. A typical pair of curves is shown in Figs. 7 and 8.

Conclusions

A technique has been evolved which appears to meet the very exacting conditions required to obtain measurements of the response of the vitreous body to external forces and of its viscous-elastic properties. This has shown that the vitreous body possesses viscous-elastic properties similar to those of gelatine, and of the same nature as dilute plasma clots. Acidification causes the gel to behave more like an ordinary elastic solid, increasing the instantaneous displacement and diminishing the "creep." The results indicate that the vitreous humour is a gel composed of a mesh-work of elastic fibrillae suspended in a viscous liquid, a view which is borne out by ultramicroscopic examination, and has previously been fully elaborated.

The technique here described could form a method for the exact study of the physical state of the vitreous gel in different pathological conditions, providing as it does an index of the consistency of the gel and its shrinkage or turgescence owing to the withdrawal or addition of water of adsorption to the colloid micellae. It is interesting that the state of a slightly acidified vitreous corresponds to that of an early degeneration and breakdown of the gel structure. Such a condition is seen in many cases of retinal detachment; and in this state the increase of sudden instantaneous displacement on the application of forces, and the diminution of the slow "creep" may be of considerable mechanical significance.

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