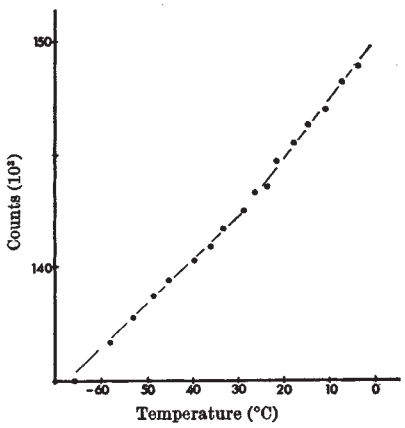

Fig. 3. Transmission of β-rays versus temperature for a sample of pure α-cellulose. Radiometric dilatometry was employed.

and $-40°$ C. The decrement value increased linearly from 0·25 to 0·30 as the temperature was decreased. The simultaneous increase in the value of the modulus was also linear.

Independent experimental evidence for the transition was obtained by a dilatometric technique based on the dependence of the transmission of β-rays on the weight/unit area of the sample at varying temperatures[5]. Because of the discontinuity of the thermal expansion coefficient in the vicinity of a transition, the transmission–temperature curves have a shape equivalent to that of curves obtained dilatometrically. The line reproduced in Fig. 3 and obtained with an improved radiometric technique shows a discontinuity slightly below $-30°$ C (unpublished work of Klason, Kubát and de Ruvo). From the slope of the two rectilinear portions of this curve, the coefficients of thermal expansion for the regions above and below the transition point were calculated to $6{\cdot}0 \times 10^{-5}$ and $4{\cdot}5 \times 10^{-5}$, respectively. The sample was prepared from bleached sulphite cellulose.

The physical background of the transition presented here is not known. Its disappearance in regenerated cellulose containing glycerol indicates that it is associated with the amorphous part of cellulose. Further work is in progress, with a view to investigating the molecular nature of this transition.

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BIOPHYSICS

Oriented Water in the Sciatic Nerve of Rabbit

A PRELIMINARY study has been made of the water found in the sciatic nerve of rabbit, which is one of the larger myelinated nerves, by proton magnetic resonance. Specimens from recently killed animals were stored in a refrigerator at about 2° C for periods of at least 1 h before use. In this process a certain amount of water was lost, as shown by condensation on the walls of the containers. Approximately cylindrical samples of the nerve, about 4 mm long and 1 mm in diameter, were examined in a Varian '*A*60*A*' spectrometer, the nerve axis being coincident with the cylindrical axis. The angle between the nerve axis and the applied magnetic field could be varied at will.

The spectra obtained are shown in Fig. 1. An appreciable amount of structure is observed and this structure depends on the orientation of the nerve within the applied field. It is consistent with the assumption that it is caused by dipolar interactions between the protons in individual water molecules. This interaction has the value $-3\mu^2/2r^3\ \overline{(3\cos^2\theta - 1)}$, where μ is the magnetic moment of the proton, r the interproton distance and θ the angle between the proton–proton vector and the applied field direction. In normal isotropic water it is averaged to zero by molecular tumbling. This experiment suggests therefore that the bulk of the water inside the nerve is in a partially oriented state.

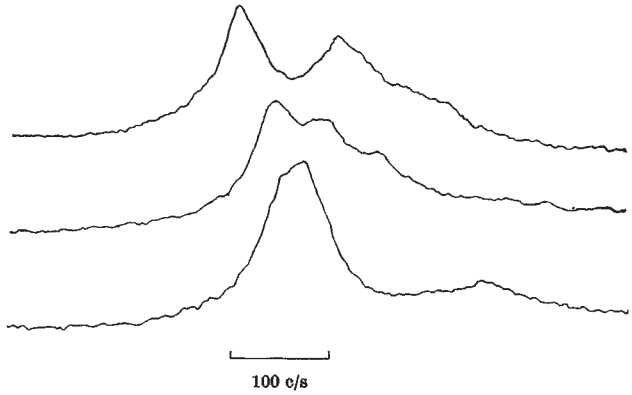

Fig. 1. The spectra of water in the sciatic nerve: (top) with nerve axis parallel to the applied field; (centre) the perpendicular orientation; (bottom) with the nerve axis at approximately 54° to the applied field direction.

The principal feature of the spectrum is a doublet the maximum splitting of which is observed when the nerve axis is parallel to the applied field. The splitting has half its maximum value in the perpendicular orientation and may be caused to be zero by setting the nerve axis at an angle of 54° 44′ with the applied field. The orientation parameter S_{22} (ref. 1) has a value of about 0·004. The movement of the centre of the doublet as the orientation of the nerve is varied is too large to be caused by a chemical shift anisotropy and must originate in the anisotropy of the magnetic susceptibility of the nerve.

The other peaks in the spectra each show orientation dependent shifts much larger than this susceptibility shift. This suggests that these also originate in partially oriented water in a different environment and in a differently oriented state from the principal component.

It is interesting to postulate possible origins of these orientation effects. There are at least four possibilities. The water may simply be constrained by Van der Waals' forces to move in annuli which must have near molecular dimensions (about 10 Å). The orientation may be secondary orientation produced on the surface of protein molecules which are themselves aligned by the pores in the nerve. Orientation may be caused by electrostatic interactions near the fibre surface. A simple calculation suggests that the field strengths feasible are insufficient to explain the orientation observed. The water could be in a chain structure as postulated for the collagen system[2]. There appears to be no justification for this explanation, and the degree of orientation observed can be explained completely in terms of normal water.

In view of the normally accepted pore sizes in nerves, which are much too large to cause orientation, it is surprising that most of the water is not in its normal state.

Further work is in progress to assess the implications of these results on the structure of myelinated nerves.

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IMMUNOLOGY

Serological Expression of Antibody Specificity for a Single Determinant of an Antigenically Complex Molecule

THE immune response to an antigenically complex molecule results in the formation of antibodies specific for one or more antigenic determinants[1]. In all but a few instances[2], antibodies specific for individual determinants cannot be identified, which precludes the evaluation of an immune response at this level of serological specificity. It is therefore usually assumed that antibodies specific for multiple determinants are regularly produced and participate in the serological reactivity of an antiserum. Such an assumption could obscure the possibility that a given antiserum consists of antibodies specific for a single determinant. The inability of such antisera to produce precipitation[3] may, in turn, lead to their being considered devoid of antibody activity or their serological properties being misinterpreted. Because of these practical considerations, we decided to describe our experience of this subject.

We became aware of antibody specificity for a single antigenic determinant during a serological examination of human tetanus antitoxin, when human sera of known toxin neutralizing activities were examined by both passive agglutination and immunoelectrophoresis. The initial examination was made by passive quantitative agglutination, a procedure previously used to compare antibody combining properties[4]. In its present application, tetanus toxoid was coupled to human group O red cells[5]. As we anticipated, there was no correlation between the magnitude of the quantitative agglutination curve and neutralizing activity (Fig. 1). Nevertheless, it was interesting that one antiserum with less than 1 U of neutralizing activity/ml. gave a strong agglutination curve. Additional examples were found among other available low-neutralizing sera (Fig. 2).

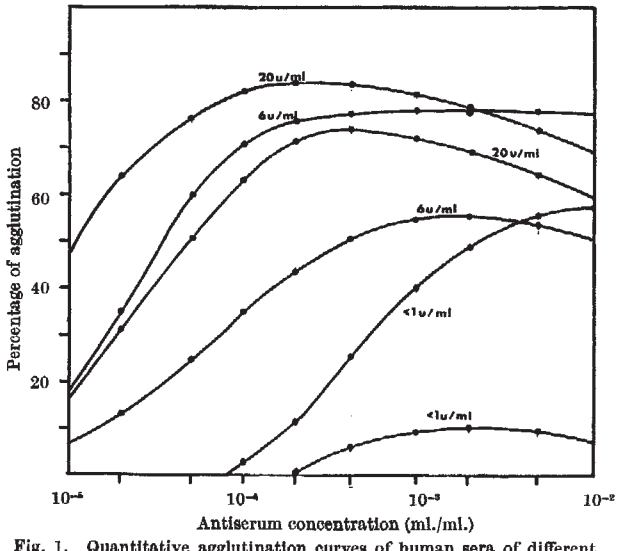

Fig. 1. Quantitative agglutination curves of human sera of different tetanus toxin neutralizing activities.

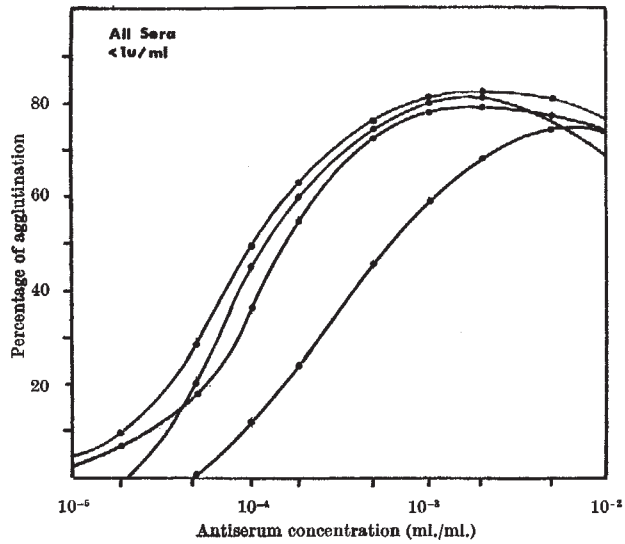

Fig. 2. Agglutinin activity of selected antisera in which little or no neutralizing activity was demonstrable.

Selected examples of both high- and low-neutralizing sera were compared by immunoelectrophoresis. All the low-neutralizing sera, and one of the high-neutralizing sera, failed to give a precipitin band. This was particularly surprising because some of the non-precipitating sera had given stronger agglutination curves than several of the precipitating sera. The failure of precipitation among the strongly agglutinating sera could not be reasonably ascribed to either low antibody concentration or affinity, and so we looked for an alternative explanation.

In comparing the mechanics of precipitation and passive agglutination, it became apparent that, although both reactions require lattice formation, antisera specific for a single antigenic determinant would behave quite differently; an antiserum specific for a non-repeating antigenic determinant would be incapable of lattice formation. It was, however, evident that if such an antiserum reacted with particles to which many antigen molecules were coupled, as in passive agglutination, then ample opportunity for lattice formation would exist. This principle, regularly considered in the assay of antibodies specific for simple chemical haptens[6], seemed directly applicable to other observations. We concluded that the strongly agglutinating, non-precipitating antisera encountered in this investigation were made up largely of antibodies specific for a single, non-repeating, antigenic determinant. Evidence supporting this conclusion was obtained later.

Two related experiments were carried out in which advantage was taken of previously demonstrated properties of individual antisera. The first experiment consisted of mixing the one available example of a non-precipitating, high-neutralizing serum (> 20 U/ml.) in varying proportions with a non-precipitating, low-neutralizing serum (< 1 U/ml.) and testing with tetanus toxoid by immunodiffusion. Neither of these sera alone produced detectable precipitation of the tetanus toxoid. When combined in equal proportions, a comparatively strong precipitin band was formed, indicating that the two sera must have been specific for two different determinants, and that both determinants were present on the same molecule. In other circumstances, the non-precipitating character of such an antiserum would undoubtedly have been misinterpreted. In the second experiment a papain digest[7] of the γ-G antibody from a strongly agglutinating, non-precipitating, low-neutralizing (< 1 U/ml.) serum was mixed with different precipitating, high-neutralizing sera in the hope of observing inhibition of precipitation with