garded as heterozygotes (Q + S +). This could be analogous to the excess of heterozygotes in the MN system among offspring of some parents who are of blood group MN (*3*).

4) In population IV, only the genes *a*, *b*, and *d* are common. The fact that the S antigen commonly occurs in combination with the Q antigen but is rarely or never expressed in its absence might lead to complicated assumptions of a biochemical or genetic nature regarding the relation between the Q and S antigens, when a simple-complex relation is assumed between pure typing reagents and antigens. This is thought to be analogous to the situation in the Rh-system, where the antigen C rarely occurs in the absence of antigen D.

5) In population V, only the genes *a*, *c*, and *d* are common. This situation is similar to the ABO-system, where (a + c +) = AB, (a + c −) = A, (a − c +) = B, and (a − c −) = O. Rare occurrence of gene *b* could, however, lead to anomalous inheritance; for example, it would be possible for an AB(*b*/*d*) × O(*d*/*d*) mating to have AB(*b*/*d*) or O(*d*/*d*) offspring.

Thus the method of classification drastically affects the interpretation of immunogenetic systems. In this report the influence of a prejudicing method of classification has partially been evaded by introducing a new code of classification. This complex-simple model is based on the introduction of a new fundamental element—the antibody specificity—which cannot be subdivided and does not cross-react. Hence the quantitative phenomenon of cross-reactivity can be discussed in qualitative terms.

Additional consequences of a theory based on the concept that antibodies consist of any combination of a restricted number of discrete antibody specificities are beyond the scope of this report.

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Muscle Volume Changes: Relation to the Active State

Abstract. *The volume of a frog sartorius muscle increases during a single twitch and subsequently decreases. The magnitude of volume change is* 10^{-5} *cm*^{3} *per gram of muscle. The time courses of change in volume and of onset of tension vary with temperature in a similar way, which implies a relation between the volume changes and the contraction mechanism. The character of the change depends upon the initial length of the muscle: the greatest decrease accompanies an isometric twitch at reference length. Treatment of the muscle with an iodide Ringer solution prolongs the active state and the volume change. These results suggest a correlation between the volume changes and the active state.*

Ernst (*1*) has demonstrated a decrease in volume of about 0.002 percent during tetany of isolated striated muscle from the frog. Meyerhof and Hartmann (*2*) showed that the decrease paralleled the development and maintenance of tension in this muscle. The occurrence of an increase in volume preceding the decrease was reported by Fischer (*3*). Most early experiments were performed with frog gastrocnemii in which tetany was induced in order to obtain an observable change in volume. However, Hill (*4*) showed that this decrease may be caused by an internal pressure of 200 mm-Hg developed within the gastrocnemius muscle.

Experiments were performed recently (*5*) with sartorius muscle, in which fibers are oriented parallel to the muscle axis, so that no volume decrease could arise from self-compression forces. The sartorius muscle is quite thin and can readily be supplied with oxygen, whereas contractions of excised gastrocnemius muscle show a noticeable deterioration because of anoxia.

In studies of the volume change in sartorius muscle during an isotonic twitch (*6*) the instrumentation used had a resolution of approximately 10^{-7} ml volume and a time response of about 1 msec. This sensitivity made muscle tetany unnecessary; the change in volume during a twitch was of greater interest than the volume change during tetany, throughout which a complex nonlinear overlap of successive changes occurred. The muscle, contained in a chamber open to the environment, was surrounded by frog Ringer solution (*7*). The solution and a wire close to a region of the solution's surface constituted two plates of a capacitor, the storage capacity of which depended upon the distance between the plates, and therefore upon the total volume of the chamber.

In the experiments reported here, we used a pressure transducer instead of a proximity transducer: the muscle chamber was closed to the environment and the change in pressure of the fluid surrounding the muscle was measured. The volume changes were quite small so that the pressure change in a closed chamber was equivalent to a corresponding volume change in a chamber at atmospheric pressure. This replacement of an isobaric system by an isochoric system resulted in a greater stability of the resting volume (recorded base line).

Measurements taken with a resolution of 10^{-7} ml contained superimposed noise introduced by high-gain amplifier circuitry and by spurious fluctuations at the transducer-fluid interface. A signal-averaging computer was employed to improve the signal-to-noise ratio of the data. Successive wave forms of volume change were digitized and added in a magnetic core memory; as the number of wave forms increased, the random noise in the input tended to average to zero, while the amplitude of the signal rose linearly. In these experiments, voltage wave forms corresponding to volume changes accompanying 16 successive twitches were averaged, with a resultant signal-to-noise ratio improvement of 4 to 1.

Sartorius muscles from *Rana pipiens* were excised and allowed to equilibrate in oxygenated Ringer solution for an hour. Muscles were removed with an attached segment of bone at the proximal end; a thread was tied to the tendon at the distal end. Muscles were mounted on a multielectrode grid system in the chamber (*8*, fig. 1). Pressure changes at the transducer, which was built into the chamber, caused a shift in the resonant frequency of an oscillator whose output was detected and amplified. The amplified output was monitored on an oscilloscope and used as the signal-averaging input for the computer. Resolution and time response of this system was also about 10^{-7} ml and 1 msec, respectively. The instrumentation for detecting changes in volume was calibrated with a small solenoid-controlled rod partially inserted into or removed from the chamber.

We considered the possibility that recorded wave forms corresponded to shock waves arising from the mechanical movement of the muscle rather than from volume changes within muscle. In several experiments a turbulence comparable to that caused by rapid muscle motion was introduced in the fluid within the chamber; this turbulence was not accompanied by a measurable volume change.

The general character and time course of the wave form for volume change against time of an isometric twitch of frog sartorius muscle was reported previously (*5*). Recently improved calibration techniques suggest, however, that the magnitude of the volume change is somewhat greater than reported. The initial volume increase is about 2.5×10^{-5} cm^3/g and the subsequent decrease about 5×10^{-5} cm^3/g.

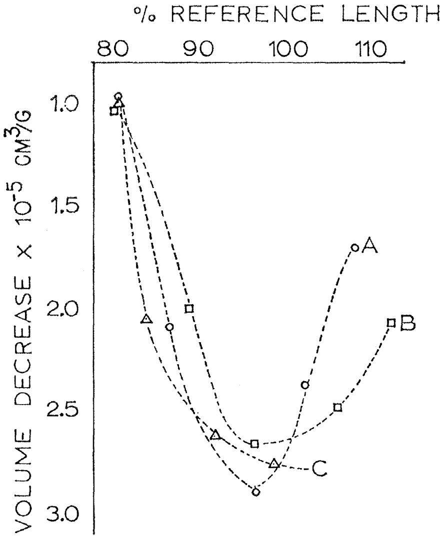

Fig. 1. The maximum volume decrease attained during a single twitch of frog sartorius muscle at 20°C is plotted as a function of the length of the muscle before stimulation. *A*, *B*, and *C* represent isometric, isotonic (moderate load), and end-free conditions, respectively.

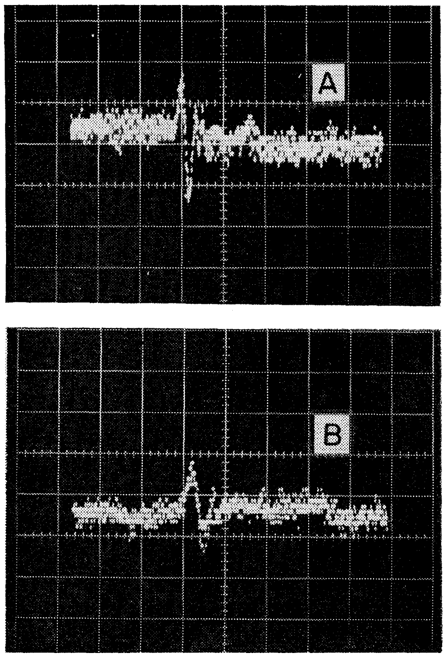

Fig. 2. Volume changes during an isometric twitch of frog sartorius muscle slightly below reference length at 25°C. (*A*) In chloride-Ringer; (*B*) after soaking in 100 percent iodide-Ringer for 30 minutes. Sweeptime, 250 msec. Amplitudes of increase and decrease are approximately 2.5×10^{-5} cm^3/g of muscle.

The magnitude of the decrease in volume is strongly dependent upon the initial length of the muscle (Fig. 1). The greatest decrease in volume accompanies an isometric twitch at reference length. This variation of decrease in volume with length of the muscle is similar to the well-known dependence on length of the tension of an isometric twitch, the tension magnitude being greatest at reference length and less at longer and shorter lengths.

The time of the onset of increase in volume, the time of maximum increase and minimum decrease, as well as the time of the onset of tension, in frog sartorius muscle are exponential functions of the temperature (*9*). The development of the increase in volume precedes any sign of mechanical activity in the muscle; tension development begins during the time of volume decrease: that is, between the times of maximum volume increase and minimum volume decrease. The volume decrease, therefore, occurs during the rise of the active state.

It is known that replacement of chloride ions by an equivalent number of iodide ions in the frog Ringer solution can affect the time course and magnitude of the twitch tension (see *10* and *11*). Iodide ions act on the excitable surface of muscle fibers to increase the duration of the active state. The increase in volume accompanying a twitch of frog sartorius muscle was of greater duration after the muscle was soaked in an iodide Ringer solution (Fig. 2). The magnitude of volume change decreased after a brief soaking; the high concentration of iodide ions caused irreversible damage to the membrane (*10*).

The effect of iodide Ringer solution upon the change in volume was threefold: although the time of onset of the increase was not affected, the rate of the rise of increase was decreased; the time at which the muscle attained maximum volume was delayed; and the time at which the minimum volume occurred was delayed. That is, iodide Ringer solution delayed the time course of the decrease in volume.

These results corroborate the earlier suggestion (*5*) of a relation between the volume changes and the active state.

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