in the tissues can be less than ambient.

Summary. The effect of the composition of the fluid in the manometer system on tissue pressure was examined. Needles were introduced into the ventricular wall of isolated, non-beating hearts. The pressure registered by manometers attached to the needles varied with the composition of the fluid in the manometer system. When distilled water was in the manometer, the pressure fell slowly to values less than atmospheric pressures; when the manometer fluid was normal saline, a positive pressure usually developed. The results are interpreted by considering that the liquid in the manometer is separated from the tissue fluids by membranes such as the walls of cells, capillaries, and connective tissue sheaths. The diffusion of water from regions of higher to lower concentrations becomes manifest in the slow development of a pressure gradient. This virtual diffusion or osmotic pressure must be differentiated from the hydrodynamic pressure which becomes manifest immediately as a pressure wave is transmitted from one compartment to another. Because the concentration of water in the tissue differs from that in the manometer, pressure recordings will represent the sum of the actual hydrodynamic pressure and the virtual osmotic pressure. Such sums may be negative with respect to the atmosphere. The manometer system thus becomes a part of the experimental design for measurement of tissue pressure.

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Electrical Conductance Changes Associated with Interaction of Viral Antigen and Its Specific Antiserum.* (31130)

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Evidence exists to suggest that electrostatic interactions play an important role in antigen antibody reactions(1). It has been shown that reactivity of antihaptenic antibodies is inhibited by ions of the same charge as the hapten(2,3). Electrical mobility value of an antigen antibody complex has also been shown to be intermediate between the values of its component proteins(4). Of additional interest is the report that maximum binding of congo red to human serum albumin is associated with a minimum in electrical conductance(5). The above suggest that reaction between antigen and antibody might also be detectable by electrical conductance measurements.

Materials and methods. Echo virus, serotype 19 and its corresponding rabbit antiserum were employed. Viral antigen was grown in monkey kidney culture tubes with Melnick monkey kidney B medium as nutrient at 37°C for 72 hours and then stored at —20°C(6). Aliquots were thawed as needed and centrifuged before use. Preimmune serum was obtained from a rabbit, after which 2 ml of antigen supernatant were injected i.v., weekly for 6 weeks. Immune serum was obtained after 8 weeks, and as-

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TABLE I. Neutralization of Echo 19 Virus* by Rabbit Echo 19 Antiserum.

Antiserum dilution	Viral growth	Volume ratio antiserum/virus
1: 20	—	50.0
1: 40	—	25.0
1: 80	—	12.5
1:160	+	6.25
1:320	+	3.12
1:640	+	1.56

* Viral preparation diluted 1:1000.

sayed for neutralizing antibodies (Table I).

For conductivity measurements, Echo 19 antigen preparation was diluted 1:1000. Its specific resistance was 23,960 ohms. Antiserum and preimmune serum were diluted 1:30. Specific resistance of these dilutions was 1,314 and 1,312 ohms, respectively. Three ml aliquots of diluted antigen were added to 2.0 ml of a mixture in varying proportions of diluted preimmune and immune rabbit sera. Ratio of antiserum/virus (AS/V) was varied from 0.0 to 22.2. This technique was adopted so that the protein environment in all tubes would be as similar as possible with only the antibody content varied. All dilutions were made just prior to use with freshly prepared deionized water ($R > 1 \times 10^6$ ohms) and placed in a thermostat (37.5°C ± 0.05°C).

Conductivity measurements were made with a General Radio Co. Impedance Bridge, Type 1608A (accuracy ± 0.1%) and a pipette type conductivity cell (cell constant = 1.18) at a frequency of 1 KC. Values are given in ohms as specific resistance (R).

Results. The initial mixture of antigen and preimmune serum (AS/V = 0) yielded R = 3,006. With addition of increasing proportion of antiserum, R dropped to 2,948 ohms at AS/V = 4.44. Thereafter, R rose to a maximum value of 3,198 ohms at AS/V = 8.89. With addition of more antiserum to a ratio of AS/V = 22.22, R decreased somewhat and tended to fluctuate between 3,184 and 3,144 ohms. These data are depicted in Fig. 1. Measurements were made 60 minutes after preparation of dilutions, but in order to determine the time dependence of the resistance measurements, one mixture (AS/V = 8.89) was measured at hourly intervals for 5 hours. The resistance showed a regular decrease of 4 ohms/hour from the initial value of 3,198 ohms.

Results of the viral neutralization test correlated with resistance data are shown in Fig. 1. The sharp resistance increase (conductance decrease) occurred within the range of AS/V ratio where the viral preparation was neutralized by antiserum.

Some of the antigen preparation was inactivated by ultraviolet irradiation† for 48 hours. The irradiated inactive antigen was then added to the preimmune and immune serum as previously described and resistance measurements were made. In the range of AS/V, 0 to 22.2, resistance readings remained constant at 3,000 ohms, and no rise in the vicinity of the neutralization point was detected (Fig. 1).

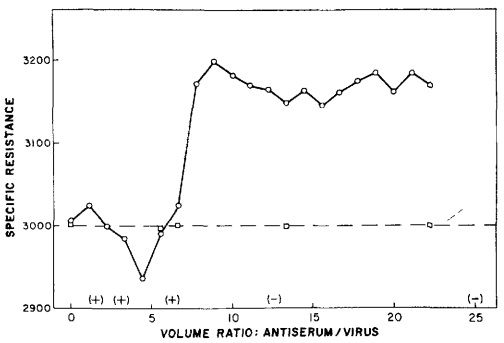

FIG. 1. Specific resistance of Echo 19 viral antigen and antiserum mixtures ○ — ○, antigen untreated, □ — □ antigen irradiated with ultraviolet light. Viral growth not inhibited (+); viral growth inhibited (—).

To test the possibility that interactions between preimmune and immune serum added to antigen might be responsible for the observed resistance changes, varying proportions of these were mixed without antigen and resistances measured (Table II). Resistance of the mixtures decreased approximately 1.5% of the resistances of either serum alone. With absence of antigen, no increase in resistance was observed.

Discussion. In these experiments, increase in electrical resistance was observed as in-

† Hanovia germicidal mercury vapor lamp, type 94A1; principal emission at 266 Mμ; distance to sample, 15 cm.

TABLE II. Specific Resistance of Mixtures of Rabbit Echo 19 Antiserum and Pre-Immune Serum Without Viral Antigen.

ml serum (diluted 1:30)		Specific resistance
Pre-immune	Immune	(ohms)
5.0	0.0	1314
4.0	1.0	1294
2.5	2.5	1300
1.0	4.0	1293
0.0	5.0	1312

creasing amounts of rabbit anti-Echo 19 antiserum were added to tissue culture preparations of Echo 19 virus. The maximum increase amounted to 192 ohms or 6.4% of the original value of 3,006 ohms in the absence of antiserum. It is noteworthy that the resistance increase was observed at antiserum concentrations capable of inhibiting growth. It appears, therefore, that the resistance increase is due to neutralization of the viral antigen by its specific antiserum. This view is supported by the fact that alterations and inactivation of the antigen by ultraviolet irradiation lead to a subsequent disappearance of the resistance changes. The reduction and alteration of antigenic specificity of protein antigens upon denaturation has been demonstrated in several systems(8).

The maximum error of conductance measurements due to instrumental and temperature effects is about $\pm$ 0.4%, or $\pm$ 12 ohms at a reading of 3,006 ohms. Thus, the maximum resistance increase of 192 ohms is some 8 times greater than possible measurement errors.

If electrostatic interactions are important in the formation of antigen antibody bonds, then interaction of unlike charges would lead to charge neutralization. Since conductance is a summation of all individual ionic conductances(9), then such a process should result in some reduction of specific conductance (resistance increase). Since antibody is protein, and since proteins can carry both positive and negative charges, this process could presumably occur even if both reactants were of the same charge type.

Summary. Measurements of electrical conductance have been performed on varied mixtures of Echo 19 virus and its specific rabbit antiserum. A 6.9% decrease in conductance (increase in resistance) was observed in the vicinity of the antibody neutralization point as measured by viral growth inhibition. No such decrease in conductance was observed with mixtures of antiserum and ultraviolet irradiated inactive antigen.

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