TABLE III. Oxygen Consumption, ml/kg/min, in Rats at a Body Temperature of 24-25°C.

Animal No.	Non-adapted rats	Animal No.	Hypothermia-adapted rats
1	15.8	5	14.3
2	17.7	6	18.5
3	22.3	7	20.0
4	23.0	8	21.4
Avg	19.7 ± 1.7	Avg	18.6 ± 1.7

$t = .46$ (non-significant)

The improved performance of the hypothermia-adapted rats probably reflects the "better condition" of the central nervous system in hypothermic animals which have previously been exposed several times to lowering of their body temperature. How much the physiological state of hypothermia-adapted animals is changed becomes evident when it is recalled that the body temperature at which adapted rats respond excellently is the body temperature barely higher than the one at which locomotion stops and a complete physical anesthesia by cold is induced.

We believe that our work shows a new type of adaptation—adaptation to hypothermia—which is measurable in terms of operant behavior at a low body temperature. After several prior exposures to hypothermia, rats cooled to a low body temperature acquire a simple technique for obtaining external heat faster and at a lower body temperature than unadapted hypothermic animals.

Summary. Hypothermic rats acquire a simple technique for obtaining external heat more quickly at a body temperature of 29-30°C than at other decreased body temperatures. When the body temperature is decreased below 25°C the latency period for acquisition of the new behavior is much extended and the performance is rarely observed during a period of 180 minutes. However, when rats are exposed to hypothermia several times prior to the response acquisition experiment they are able to respond steadily not only at a body temperature of 25°C but even at temperatures between 22° and 25°C. While all hypothermia-adapted animals thus responded at these body temperatures (22.2-24.7°C), 5 out of 7 non-adapted animals failed to do so. The performance latency time of the 2 non-adapted rats which eventually performed was longer than that of any hypothermia-adapted rat. It is concluded that previous exposure to hypothermia ("adaptation to hypothermia") enhances the response acquisition ability of rats at low body temperatures.

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Received March 7, 1966. P.S.E.B.M., 1966, v122.

The Manometer Factor in Measurement of Tissue Pressure.* (31129)

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Tissue pressure has been reported on occasion to be negative with respect to atmospheric pressure(1-3). Guyton(2,3) implanted small perforated plastic balls in the subcutaneous connective tissue; after subsidence of the acute inflammatory process, subatmos-

* This study was aided by USPHS Grant HE 08721 from Nat. Heart Inst.

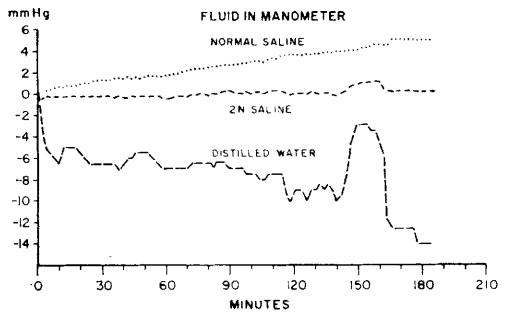

FIG. 1. Manometer readings of "tissue pressure" in a non-beating heart. Vertical axis is in mm Hg. Horizontal axis is in min. All readings begin at atmospheric (zero) pressure. Upper (dotted) line is pressure recorded in a manometer which contained 0.9% (normal) saline. Central line is recorded pressure obtained with the manometer filled with twice normal (1.8% NaCl) saline. Lowest line is pressure tracing obtained when the manometer system was filled with distilled water.

pheric pressures were recorded in manometers connected to needles inserted into such perforated balls.

Since our concept concerning the regulation of vascular conductance is dependent on a positive tissue pressure which partially or completely collapses the capillaries(4), we have examined previously neglected aspects of the measurement of the tissue pressure.

Methods. Five isolated, non-beating dog hearts, and 3 differential transformer manometers (Sanborn 267B) were used.

Heart. Three needles (#25 gauge) were introduced into the left ventricular wall of each isolated heart in a manner such that all the tips were on the same horizontal plane. Pressures were recorded for periods up to 5 hours. In 2 hearts, pressures were recorded on 3 successive days.

Manometer. The manometer tubes were filled with either (1) distilled water, (2) half normal saline (0.45% NaCl) solution, (3) normal physiological saline (0.9% NaCl), or (4) twice normal saline (1.8% NaCl). Tracings were made on direct writing paper.

Results. The pressures recorded from the needles varied with the composition of the fluid in the manometer system (Fig. 1). In the manometer filled with distilled water, the pressure fell below atmospheric levels and moved irregularly, registering values as low as —14 mm Hg. A positive pressure usually developed in the manometer system containing normal saline. When the fluid in the manometer was either half or twice normal saline, the pressure usually fell at first; it then fluctuated in an irregular manner over the course of the 3 hours or so of each experiment. Similar results were obtained on hearts left at room temperature and tested on 3 successive days.

Discussion. Measurements of tissue pressure in the living animal are subject to errors because of local hemorrhage, trauma, edema, or disturbances in ions or diffusion patterns (5). Thus, insertion of a fine needle into a tissue will almost invariably traumatize some capillaries and other small blood vessels; the extravasation of blood into the tissues generates a positive pressure which, in enclosed compartments, may approach arterial values.

Even if the hemorrhage could be obviated, local anatomy and physiology are disturbed by distortion and displacement, modifying the local tissue pressure. Even minimal local trauma, which appears to be negligible, can cause serious disturbances in water binding and local transient edemas may develop. This has been shown especially in the skin(6).

Manometers used for measurement of tissue pressure ordinarily contain a variant of "physiological" saline, such as Ringer's solution. The compositions of these solutions have been based on studies of the electrolyte patterns in the blood plasma. However, concentrations of ions, proteins, and fluids in the tissue spaces probably differ greatly from those of the blood. The ultrafiltrate of the blood which leaves the capillaries to enter the extravascular spaces is quickly modified by the extraction of substrates and oxygen by the parenchymal cells, and by their contributions of carbon dioxide, potassium, and other materials. While saline in the manometer needle may be properly balanced against the ions of the blood, there is no guarantee that such a solution is in "physiological" balance with the tissue fluids. A proper balance requires that the fluid in the manometer have the same composition as that of the fluid in the tissue spaces of the organ under examination.

Substances diffuse from regions of higher

to lower concentrations(7-9). Thus, water of high concentration (distilled water) will diffuse into an adjacent body of water which is diluted by solutes. When a membrane separates 2 compartments which contain waters of unequal concentrations, the tendency to diffusion will be registered as an "osmotic pressure." Thus, the diffusion of the concentrated water of a manometer system across a membrane into the diluted water of a tissue lowers the pressure in the manometer. The process is slowed by the membrane and the pressure difference develops slowly. This is evident clinically in the tendency for fluid slowly to enter and enlarge subdural hematomas, thereby distorting and injuring the brain.

The membranes of the capillaries, cells, and connective tissues separate the water in the manometer system from that in the tissues. When the needle of a manometer is inserted into a tissue, the relative concentrations of water in the manometer system and in the tissue will therefore produce a pressure reading which differs from ambient values.

The present results confirm the findings of Guyton(2,3) that insertion of a needle into a tissue may result in the gradual appearance of a subatmospheric pressure in an attached manometer. However, recorded negative pressure in such a circumstance does not represent a true subatmospheric hydrodynamic pressure. Instead it measures a *virtual* pressure due to the forces of diffusion.

Authoritative evaluations of diffusion and osmotic pressure equilibria have been presented by a number of workers(7-9). Less attention has been given to differences between dynamic pressures and those which result from diffusion.

The hallmark of the passage of a pressure probe such as a needle across a retaining wall into a container with a different hydrodynamic pressure is the registration of the new pressure within a few milliseconds. By contrast, when a needle or a cannula of a manometer is separated from a compartment by a semipermeable membrane, the diffusion of water across the membrane becomes manifest as a much slower change in pressure, as seen in the reports of Guyton(2,3).

The delay of seconds or even minutes in the establishment of an equilibrium in measurements of "tissue pressure" indicates that this effect must be attributed to diffusion rather than to a hydrodynamic force. The rate of achievement of equilibrium is then determined by the relative concentrations of water in the compartments separated by the membranes, the surface area across which the diffusion takes place, the porosity of the membrane, the compliance of the manometer system, and any differences in temperature of the two systems.

Numerous workers have utilized equilibration pressures produced by injecting "physiological" solutions into a tissue and then waiting for the pressure to fall to a stable level. Such injections disturb the normal state of the tissue and thereby may introduce artifacts into the measurement. The oscillations in pressure which we observed in the muscle experiments probably represent changes in the concentrations of the water as the concentrations of solutes change in the membranes and fluid. In a simple model, the change in pressure follows a logarithmic decay curve; similar logarithmic traces are evident on inspection of published data on "tissue pressure"(1-3).

A negative hydrodynamic pressure cannot be maintained in a flexible container unless a pump is available to remove the fluid at a rate faster than the hydrodynamic forces can deliver it. Since the skin is a flexible container, a local negative tissue pressure must necessarily result in the hydrodynamic inflow of water from adjacent sites, or in the collapse of the tissue by the higher atmospheric pressure. The maintenance of a negative pressure in the face of the difficulties posed by the flexibility of the integument of mammals and birds would thus demand a remarkable and continuous energy expenditure.

The present analysis calls attention to the need clearly to separate the hydrodynamic pressure which appears immediately as unimpeded flow takes place between a manometer system and a compartment, from the virtual pressure generated by diffusion of water between these systems. While negative pressures may be recorded from tissues, it is unlikely that the hydrodynamic pressure

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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Received October 4, 1965. P.S.E.B.M., 1966, v122.

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-

* Supported in part by Grant AM 06380 from the USPHS and by a grant from New York State Chapter, Arthritis Foundation.