THE PRESSURE AND INTERSTITIAL RESISTANCE PREVAILING IN THE NORMAL AND EDEMATOUS SKIN OF ANIMALS AND MAN

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Knowledge of the pressures existing within tissues is essential to an understanding of lymph formation and fluid exchange. Pressure within a tissue, when it exists, must tend to hinder the escape into it of fluid from the blood and thus affect lymph flow. In an attempt to procure some needed knowledge concerning the state of affairs within cutaneous connective tissue, we have employed methods recently developed in this laboratory (1–3) for the measurement of fluid pressure in edematous skin and for the study of the resistance of normal and edematous cutaneous tissues to the introduction of minute amounts of fluid. The experiments which will be presented here have been carried out upon the skin of the ears, backs, and thighs of mice, and that of the arms and legs of men. As yet, no reason has been found to suppose that the findings apply to tissues of other sorts.

In the past when workers have attempted to determine the pressure within cutaneous tissues, the minimum pressure required to force small amounts of physiological saline solutions into normal skin has been taken as implying that a nearly equivalent tissue tension or pressure prevailed. Few investigators have realized that the amounts of fluid they employed have been great enough, as a rule, to force the tissue elements apart and set up artificial pressures. When edema develops under conditions of disease, this is what happens and it is possible to measure the interstitial pressure with certainty by determining the pressure of the extravascular fluid directly. In normal cutaneous tissue though, there is not enough freely movable interstitial fluid to allow one to make even micromanometric determinations of pressure. Those who have recently observed living intracutaneous tissue by micro methods (4–6) have been impressed by the evidence that normally it contains no open spaces, such free fluid as may be present seeming to exist in thin films on the surfaces of the formed structures instead of lying in pools between them. The nearest one can come to measuring directly the interstitial pressure under such conditions is to introduce into the tissue the least possible amount of an unabsorbable fluid that will serve as an indicator and then to determine the lowest pressure that will cause the slightest measurable movement inwards of the fluid against the resistance of the tissues. In order to avoid the creation of artificial pressure the movement

should be so slow that distortion of structure is minimal or absent. When this is the case the pressure required to overcome the interstitial resistance should not be very different from the true interstitial pressure, and without measuring the latter directly one should be able to estimate it with sufficient accuracy for most practical purposes.

In work already reported (1–3) experiments have been performed in which exceedingly minute amounts of fluid were introduced into the skin through the smallest hypodermic needles in such a manner that neither blood nor lymphatic capillaries were entered directly (1). Under these circumstances, an absorbable fluid, Locke's solution, brought into contact at atmospheric pressure with the connective tissue of the skin of an anesthetized mouse entered it intermittently at the average rate of 0.06 c.mm. in 5 minutes, withvariations, as a rule, between 0.04 and 0.08 c.mm. For this inward flow it had somehow to pass through the tissues from the tip of the injecting needle to the nearest draining vessels at least, and since there was no pressure gradient there could scarcely be any displacement or distortion of the formed elements. An unabsorbable fluid such as mineral oil, brought into contact with the tissues under similar conditions, did not enter them, and homologous serum, which was relatively unabsorbable in comparison with Locke's solution, generally failed to enter. Of more interest for the present work was the finding (1) that Locke's solution, when mixed with only 1/4 to 1/2 per cent of a blue vital dye, pontamine sky blue, no longer entered the tissues when introduced into them at atmospheric pressure. The dye mixture, although its viscosity is like that of plain Locke's solution, behaved instead like an unabsorbable fluid. The reasons for this behavior have been discussed in earlier papers (1–3, 5, 6).

These findings suggested that the criteria discussed above for an approach to the measurement of pressure in the skin might be approximately met by using the dye-Locke's solution mixture to determine the lowest pressure that will produce the least measurable movement of the fluid against the tissue resistance. With the apparatus here employed the least movement inwards of fluid that could be measured with accuracy happened, by coincidence, to be of about the same magnitude as that at which plain Locke's solution passes into skin when under no pressure (1). Accordingly, this rate, of 0.06 c.mm. per 5 minutes, was adopted as an arbitrary standard for the measurement of tissue resistance and the pressure required to maintain the standard rate of inflow constant under differing conditions will be termed the *interstitial resistance*. It is to be clearly recognized that the interstitial resistance is not a measure of the actual pressure in the tissues and that it must necessarily be slightly higher than the latter. Nevertheless, changes in true pressure within the tissues should be thrown into sharp relief.

Attempts at direct measurement of the pressure within tissues were first made in 1884 by Landerer (7) in rabbits, dogs, and man. After introducing saline solutions

at atmospheric pressure into the subcutaneous tissues, pressure was put upon the introduced fluid until it entered the tissues in perceptible amounts, which were not measured. The pressure was then lowered until inflow ceased and the resulting measurement was considered equal to the pressure sought. Landerer reported, for all the species just mentioned, pressures averaging 5.0 to 7.0 cm. of water, but in a few instances they ranged up to 60.0 cm. of water.

Forty-three years later, Hajen (8) attempted a study of intracutaneous pressure conditions by measuring the pressure required to inject relatively large amounts of saline solution, 0.01 to 0.02 cc., into the skin with force great enough to produce wheals. He recognized that the method involved an artificial pressure sufficient to burst the tissues apart. For normal human skin the wheal-forming pressure was found to be as much as 100 to 200 mm. of mercury. In patients with cardiac edema, lower pressures of 55 to 70 mm. sufficed, as did a pressure of 50 mm. of mercury during periods of circulatory obstruction in normal limbs. In these experiments the disruption of the tissues was so great and the amounts of fluid injected were so large that the resulting pressures found had no relation to the existing intracutaneous pressure but represented merely changes in the tensile strength of the tissues to mechanical traumata.

Attempts to measure actual intracutaneous pressures were first made by Meyer and Holland (9, 10). These authors stressed the fact that large amounts of an absorbable fluid can be run into the tissues very slowly, as during hypodermoclysis, and disappear there without much pressure change. To obtain change that could be measured, they injected saline solution into the skin of men at high pressure, about 70 cm. of water, with result that it entered the tissues at the rate of 70 c.mm. a minute. After flow had begun the pressure was rapidly lowered until the rate became 10 c.mm. a minute. Plotting the measurements of pressure and flow, they determined by extrapolation the point at which presumably no flow would occur. The indicated pressure, which seemed to range between 5 and 9 cm. of water in normal skin, was considered by them to represent the tissue pressure. The assumption is open to two serious objections, —too much fluid was introduced and at too high a pressure. As shown in recent work from this laboratory (1–3), and discussed above, fluids forced into the skin of mice at low pressures (1.5 to 2.5 cm. of water), moved interstitially at the average rate of only 0.06 c.mm. in 5 minutes. The inflow obtained by Meyer and Holland, through a Pravaz cannula, presumably of about the same size as the needle used by us, would seem to have been 166 to 1162 times as much. Furthermore, as bearing on the pressures employed by these authors, our work (3) has shown that fluids, irrespective of their character, when introduced interstitially into skin under gradually increasing water pressures, show a sudden acceleration of inflow when it becomes 8.5 cm. or more. The evidence indicated that this happens because the tissues are broken apart and the occurrence was termed the "breaking point." Even under these circumstances the rate of inflow at pressures between 8.5 and 20 cm. of water was less than 0.2 c.mm. in 5 minutes. It is clear that in the work of Meyer and Holland the high pressures they brought to bear at the beginning of each experiment far exceeded the "breaking point" (3) and that their measurements, like those of Hajen, were made in tissues that had been forced apart by the production of an interstitial bleb. Under these pathological conditions of tissue disruption, pressures less than normal were

found in states of generalized edema. As will be seen below, improved methods disclose an opposite state of affairs. It is to be noted, however, in relation to the data to be presented that Meyer and Holland's measurements were made long after edema had formed, under which circumstances the interstitial pressure may have returned approximately to that of the normal skin.

Wells, Youmans, and Miller (11) introduced saline solution into human skin through a fine needle, varying the pressure until there was neither inflow nor outflow. They do not give the amounts of fluid injected or the highest or lowest pressures used for making the determinations. They found intracutaneous tissue pressures lying in the range reported by Meyer and Holland, that is to say, between 5 and 9 cm. of water. Prolonged congestion in the leg from standing increased these values to 7.0 to 12.5 cm. of water, and venous congestion increased them by 2 or 3 cm. of water. Subcutaneous tissue pressures between the values of 2 to 6 cm. of water usually rose to values of 11 and 15 cm. of water in tissues overlying the gastrocnemius and anterior tibial muscles of a subject who had been standing for 3 hours.

Burch and Sodeman (12, 13) determined subcutaneous tissue pressures in man by an improved method requiring not more than 0.5 c. mm. and usually but 0.1 c.mm. of saline solution, which was introduced through a gauge 26 needle connected with a glass adapter 1 mm. in bore. In different areas of skin, at heart level, mean pressures of 1.79 to 3.71 cm. of water were found, with extremes of 0.8 to 5.4 cm. of water. Quiet standing increased the subcutaneous tissue pressure in the leg from 5.37 cm. of water, on the average, to 8.05 and in one instance to 10.2 cm. In conditions of cardiac edema, pressures in the subcutaneous tissue of the pretibial regions varied from 4.7 to 26.2 cm. of water, and decreased as edema decreased. These authors report no studies of intracutaneous pressures.

Other investigators (14–25) have estimated tissue pressure or tissue tension by indirect methods. Their work will be considered in a following paper.

It is worthy of comment that workers who have introduced small amounts of fluid into the tissues have found the tissue pressure lower than those who have injected larger amounts. The work of Burch and Sodeman, who employed the smallest amounts of fluid utilized, approached nearest to physiological conditions. In several of the techniques described the insertion of large needles into the tissues has no doubt ruptured blood vessels and lymphatics and the introduced fluid may have entered them directly.

It is clear that the earlier workers have generally introduced fluids into the tissues in far greater amounts than we employed in the experiments here to be described, consequently greater tissue resistances were encountered by them. We shall follow the terminology of these authors and designate the pressure found as *tissue pressure* whenever the occasion arises to discuss the studies in other laboratories of the pressure within tissues as obtained by the introduction of considerable amounts of fluid. The term *interstitial resistance* will be used only in connection with the measurements made in the present work. The term *interstitial pressure* will be reserved for the true pressure. This cannot be measured in normal skin by any method thus far devised. Earlier workers

have occasionally measured the pressures of edema fluid without attempting to introduce additional fluid into the tissues, and in the present work, we have made similar measurements. The edema fluid pressure found under these circumstances measures, of course, the true interstitial pressure then prevailing and the terms can be used interchangeably in discussing either the present findings or those of preceding authors.

The Interstitial Resistance in Living Mouse and Rabbit Skin

Methods.—In previous work from this laboratory (1, 2) small amounts of certain test fluids have been brought into contact with the tissues of the skin in such a manner that they enter neither blood vessels nor lymphatics directly. A 30 gauge hypodermic needle filled with a test fluid was inserted into a minute tunnel in the skin formed just beforehand by a dissecting needle of much finer dimensions. The hypodermic needle was connected with a horizontally placed graduated pipette containing the test fluid. The movement of the meniscus of the latter in the pipette, observed through a microscope and measured with the aid of micrometer eyepieces, indicated the entrance of fluid into the tissues. The apparatus was submerged in a constant temperature bath in order to measure with accuracy the minute volumes of fluid dealt with.

In the present experiments the same method was used and two test fluids employed: one, a mixture of Locke's solution with ¼ to ½ per cent of a colloidal vital dye, pontamine sky blue; the other, homologous serum. The dye-containing solution induces, in about half an hour, a mild edema which increases slightly the bulk within the tissues (2, 3); serum, as is well known, is absorbed from the tissues very slowly.

To determine the interstitial resistance one or the other of the test fluids was brought, at atmospheric pressure (1–3), into contact with the dermal connective tissue of the ears, backs, or thighs of mice and of rabbits anesthetized with luminal or nembutal. The meniscus of the test fluid in the injecting pipette was watched. After a period of 10 to 15 minutes, if no movement of the meniscus occurred, a pressure of 0.5 cm. of water was brought to bear upon the fluid in the pipette and thereafter the pressure was raised every few minutes by small increments until flow began. Pressure was then held at the height necessary to maintain an inflow of 0.04 to 0.08 c.mm. per 5 minutes, which corresponds with the speed at which Locke's solution enters the skin at atmospheric pressure. Flow at this rate resulted from pressures averaging 1.7 cm. of water.

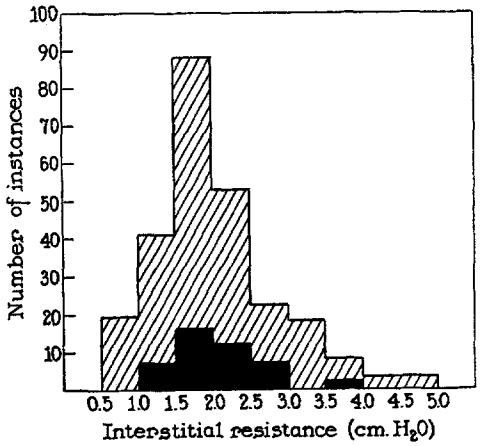
In a few exceptional instances, the dye-Locke's solution entered the tissues in the absence of any extra pressure upon it, inducing within 15 to 20 minutes an edema which resulted in backflow of the fluid into the apparatus. Further studies of such instances were abandoned, except where measurement of the pressure of the edema fluid was desired.

In a few of the experiments in which serum was used as a test fluid, it entered the tissues when under no pressure for reasons that are not understood. These instances, too, were discarded. In the remainder the course of events was like that in the experiments in which the dye-Locke's solution was used.

Control Observations on the Skin of the Mouse.—The results of more than 250 determinations of the interstitial resistance of the normal skin of the ears, backs, or thighs of mice anesthetized with luminal or nembutal are presented in Text-fig. 1. The hatched columns indicate the number of instances in which the intradermal interstitial resistance was found between 0.5 and 1.0, 1.0 and 1.5, 1.5 and 2.0 cm. of water, respectively, and so on up to 5.0 cm. of water, the high-

est readings obtained in normal skin. In these instances the measurements were made with the dye-Locke's solution. By far the greatest number fell between 1.5 and 2.0 cm. of water, which can be taken as the normal interstitial resistance of the skin of the mouse. Only rarely were resistances found as high as 4.5 to 5.0 cm. of water. The black columns in Text-fig. 1 represent the results of 44 determinations of interstitial resistance made with homologous serum instead of with the dye-Locke's solution.

In all these tests no obvious differences were found in the interstitial resistance of the various regions of the skin. This was to be expected, for the skin was loose in the areas chosen for experiment. In work to be reported later, we



TEXT-FIG. 1. The interstitial resistance in the normal skin of the ears, backs, and thighs of mice. (See text.)

have found a higher interstitial resistance in the tense, tough skin near the feet of these animals.

In 10 experiments the interstitial resistance was measured in the ears of animals killed with chloroform 1 to 4 hours before.[1] In 6 of the 10 instances, the resistance was found to be slightly less than 2.0 cm. of water; in the remaining animals measurements of 2.5, 2.5, 3.0, and 4.0 cm. were obtained.

The Effect of Hyperemia.—In an earlier paper (2) we have shown by means of the injection apparatus employed for the present work that the absorption of Locke's solution at atmospheric pressure by living skin is increased temporarily when active hyperemia occurs, in spite of a dilatation of the vessels which might tend to force the fluid, already introduced into the tissues, back into the injection apparatus. What of the interstitial resistance of the tissues under these conditions?

[1] Experiments on the skin of dead animals were not undertaken for at least an hour after death, to avoid errors that might be caused by temperature changes in the ear.

In each of twenty mice the intradermal interstitial resistance was determined in one ear. As soon as this had been done, the beam of an arc light was played upon the animal's other ear or upon its back, or both, with result that a reflex hyperemia occurred in the ear under test (2), which was kept shaded. The degree of this hyperemia varied, being sudden and intense in some of the animals and moderate in others. Prior to the use of the light, determinations of interstitial resistance varied from 1.0 to 2.6 and averaged 2.0 cm. of water. During the first 10 minutes of hyperemia the interstitial resistance rose but little or not at all and never exceeded 4.0 cm. of water.

After 15 minutes the hyperemic skin surrounding the needle usually became perceptibly edematous, as observed under the microscope. The findings when this had happened will be described further on.

No significant changes of interstitial resistance were found in hyperemic skin prior to the onset of edema.

Changes Occurring in the Edema Fluid Pressure and in the Interstitial Resistance in Edematous Skin: Relationship between the Two

When skin becomes edematous, edema fluid is generally, but not always, sufficiently free in the tissues to permit direct measurement of its pressure. If the needle of an injecting apparatus, already filled with a test solution at atmospheric pressure, is inserted into the skin, flow will usually occur from the tissues to the apparatus. The pressure required to stop this flow measures the pressure of the edema fluid which, of course, equals the interstitial pressure.

The findings of others, summarized earlier in this paper, show little agreement concerning the pressure conditions in edematous skin. Since in previous studies (23, 24), various ways had been found to induce edema in the skin of mice and to recognize its presence without resorting to "pitting on pressure" or otherwise disturbing the tissue, the subject has been studied anew, with an added aim, to determine the relationship between interstitial resistance and interstitial pressure in edematous skin.

To attain this end, we induced in the skin of many mice, by means to be described, an obvious edema, severe enough to be recognized by observation only, and we determined in the edematous skin, under a variety of conditions, both the edema fluid pressure and the interstitial resistance. In some experiments the interstitial resistance was first measured in normal skin, which either became edematous later during the experiment or was rendered so by the application of irritants. In other experiments the edema fluid pressure and interstitial resistance were measured at various intervals after edema had been produced experimentally. As will appear from the data to be presented, the interstitial resistance was found, on the average, to be about 0.5 cm. of water higher than the edema fluid pressure.

The Absence of Free Fluid in Certain Cutaneous Edemas.—In some instances in the course of these studies, a striking phenomenon appeared: free fluid could not be demonstrated in the edematous skin. Fluid failed to flow into the injection apparatus not only when the pressure in it was atmospheric but even

when mild suction was applied and after it had been ascertained by means of a plunger (1) that the needle was not obstructed. The phenomenon will be considered in later work. Suffice it to say here that in such instances we were able to determine only the interstitial resistance of the edematous skin.

TABLE I

Intradermal Interstitial Resistance and Edema Fluid Pressure during the Formation of Edema in Hyperemic Ears (See Text)

1	2	3	4	5	6
Exp. No.	Initial interstitial resistance	Interval after appearance of edema	Edema fluid (interstitial) pressure	Subsequent interstitial resistance	Remarks
	cm. of water	*min.*	*cm. of water*	*cm. of water*	
		10	—	5.5	Backflow against pressure of 4.0
		20	—	6.5	" " " " 5.5
1	2.2	30	7.0	7.5	" " " " 6.5
		60	8.5	9.2	" " " " 8.0
		90	8.5	9.0	" " " " 8.0
		10	—	4.5	
		30	4.5	4.8	
2	1.7	60	6.8	7.0	Backflow against pressure of 6.0
		90	6.8	7.2	" " " " 6.0
		10	—	3.0	
		20	3.0	3.5	Backflow against pressure of 2.5
3	2.5	30	4.2	4.5	" " " " 4.0
		80	4.5	5.0	" " " " 4.0
		10	—	2.0	
		20	2.0	2.5	Backflow against pressure of 1.5
4	1.9	30	—	4.1	
		60	—	5.8	
		90	5.5	6.2	" " " " 4.5
		15	—	2.2	
		30	—	3.5	Backflow against pressure of 2.2
5	2.2	60	—	4.0	
		90	3.5	4.0	" " " " 3.0

Changes in Edema Fluid Pressure and in Interstitial Resistance in the Skin during the Formation of Edema

As already mentioned, active hyperemia induced in the ears of 20 mice had, during the first 10 minutes of its appearance, no significant effect upon the interstitial resistance. However not infrequently the retention of the injecting

needle in the hyperemic skin for longer periods led to the development of edema. In 10 instances in which this occurred the edema fluid pressure and interstitial resistance were determined, from time to time, as edema formed.

The findings from 5 experiments are summarized in Table I. In all of them the interstitial resistance was first determined in normal skin of the ear and immediately thereafter hyperemia was induced. In all, the edema became visible after about the same time interval following the induction of the hyperemia, that is to say, after approximately 15 minutes. The table shows the interstitial resistance as first determined in the normal skin (column 2) and its subsequent changes (column 5), together with the pressure of the edema fluid (column 4), after various intervals following the first recognition of edema by the observer. The speed at which the edema developed and its intensity varied greatly from animal to animal. As there is no satisfactory way to express the degree of edema quantitatively, the data from the five instances, given in Table I, are arranged in the order of apparent rapidity of occurrence as judged arbitrarily by the observer.

In all the experiments the edema fluid pressure and interstitial resistance rose as edema developed and reached 4.0 to 9.2 cm. of water within an hour after the edema first became perceptible. In all, the edema fluid flowed into the injection apparatus whenever the pressure in the latter was less than that of the edema fluid. The highest interstitial resistance and edema fluid pressures were found in those instances in which edema developed fastest, those which have been placed at the top of Table I.

Intracutaneous Pressure Changes Following the Topical Application of an Irritant Fluid

Changes during the Formation of Edema.—In 14 experiments the needle of the injecting device was placed in the skin of the ear and the interstitial resistance was measured. Next the ears were lightly painted once with xylol and almost immediately an intense hyperemic flare made its appearance and edema developed. Pressure measurements were made at intervals during the formation of edema.

In 11 of the 14 experiments, there was backflow of edema fluid into the injecting apparatus when the pipette was opened to the atmosphere after edema had developed. In the remaining 3 instances no backflow occurred. In all, the intradermal interstitial resistance was found to be increased. The data from the experiments are arranged in Table II in the order of the speed of visible edema formation, the fastest instances at the top, as in Table I. It will be noted (columns 4 and 5) that the edema fluid (interstitial) pressures and interstitial resistances recorded in the upper part of the table are greater in general than those appearing in the lower portion.

In these experiments the edema fluid pressure and the interstitial resistance increased during the formation of edema, at times markedly, at times but little. This was to be expected, for the ears, as seen under the microscope, showed clear differences in the intensity of the edema and in the reaction of the blood vessels to the stimulus of a single painting with xylol. The more rapidly the edema formed, the greater was the resulting edema fluid pressure and the interstitial resistance.

TABLE II

Changes in Intradermal Edema Fluid Pressure and Interstitial Resistance in the Ear of the Mouse during the Formation of Edema Following the Application of Xylol to the Skin

1	2	3	4	5	6
Exp. No.	Initial interstitial resistance	Interval after painting ear with irritant	Edema fluid (interstitial) pressure	Subsequent interstitial resistance	Remarks
	cm. of water	*min.*	*cm. of water*	*cm. of water*	
1	1.0	10	—	2.8	Backflow against atmospheric pressure
		20	3.0	3.5	—
		30	4.0	4.5	—
		60	5.5	6.0	Backflow against pressure of 5.0
2	1.8	15	—	2.0	Backflow against atmospheric pressure
		30	4.0	4.5	—
		45	—	6.0	—
		60	8.5	9.0	Backflow against pressure of 8.1
3	1.5	20	1.7	2.0	Backflow against atmospheric pressure
		33	2.5	3.0	—
		40	—	3.5	—
		50	—	5.7	—
		60	9.1	10.0	Backflow against pressure of 8.4
4	3.0	10	—	3.5	
		20	—	4.5	No backflow
		30	—	4.5	
		60	—	5.0	
5	2.2	10	—	2.5	Backflow against atmospheric pressure
		20	—	3.5	" " pressure of 2.5
		30	3.6	4.2	" " " " 3.3
		45	—	5.5	—
		60	5.3	5.8	—
6	1.9	15	—	3.5	
		30	—	6.0	No backflow
		45	—	3.5	
		60	—	3.5	
7	1.5	15	—	2.5	—
		20	—	4.0	—
		30	5.0	6.0	Backflow against pressure of 4.5
		40	—	5.0	—
		45	3.0	4.0	—
		60	3.5	4.0	Backflow against pressure of 3.3
8	2.0	10	—	2.0	Backflow against atmospheric pressure
		25	2.0	2.5	—
		45	3.5	4.0	—
		60	4.0	4.5	Backflow against pressure of 3.3
9	1.7	15	—	—	Backflow against atmospheric pressure
		30	—	3.4	—
		45	—	4.5	—
		65	5.0	6.0	Backflow against pressure of 4.0

TABLE II—*Concluded*

1	2	3	4	5	6
Exp. No.	Initial interstitial resistance	Interval after painting ear with irritant	Edema fluid (interstitial) pressure	Subsequent interstitial resistance	Remarks
	cm. of water	*min.*	*cm. of water*	*cm. of water*	
		10	—	3.0	
		15	—	4.0	
10	2.0	30	—	6.0	No backflow
		45	—	4.0	
		60	—	4.0	
		30	3.0	3.5	Backflow against pressure of 2.0
11	2.0	45	—	4.5	—
		60	3.8	4.3	Backflow against pressure of 3.5
		20	—	2.0	Backflow against atmospheric pressure
12	1.7	45	—	3.0	—
		60	3.0	3.5	Backflow against pressure of 2.6
		20	—	2.0	—
		30	2.1	2.5	Backflow against pressure of 1.8
13	1.5	45	—	2.5	—
		60	2.0	2.8	Backflow against pressure of 1.5
		15	—	2.3	Backflow against atmospheric pressure
14	2.1	30	—	2.5	—
		60	2.5	3.2	Backflow against pressure of 2.0

Later Changes.—Pressure conditions in the skin vary much after the formation of edema and during its absorption. In the preceding experiments measurements were carried out for only an hour after the application of xylol. To learn something about the pressure changes in the skin at longer intervals after the original application of an irritant it seemed necessary to make measurements in another series of animals, to avoid complications arising from the added irriration resulting from too long contact of the injecting needle with the skin.

The ears of 30 normal mice were painted with xylol and thereafter, at intervals varying from 1½ hours to 11 days, individuals were selected at random for study. The interstitial resistance in the ears was measured in all instances and compared with the edema fluid pressure in all those which showed the presence of free edema fluid.

The findings are summarized in Table III. In 11 experiments performed within 1½ to 6 hours after the induction of edema the interstitial resistance was found higher than normal in only 3 (Experiments 1, 3, and 11). In 10 experiments made 19 to 24 hours after inducing edema, the resistance was high in 5 (Experiments 13, 15 to 18), and slightly higher than normal in 3 (Experiments 14, 20, and 21). In 2 experiments done after an interval of 2 days (Experiments 22 and 23) the interstitial resistance was high. In 7 studies made at longer intervals no increase was observed.

The rise of interstitial resistance found 19 to 24 hours after painting the ears with xylol

TABLE III

Intradermal Edema Fluid Pressure and Interstitial Resistance in the Ear of the Mouse after Formation of Edema and during Its Absorption

1	2	3	4	5
Exp. No.	Interval after painting with xylol	Edema fluid (interstitial) pressure	Interstitial resistance	Comments
	hrs.	*cm. of water*	*cm. of water*	
1	1½	5.5	6.0	Backflow against pressure of 5.1
2	1¾	1.0	1.5	" " atmospheric pressure
3	3	—	10.8	—
4	3½	1.0	1.5	Backflow against atmospheric pressure
5	3½	—	2.0	No backflow
6	4	Not measurable	1.8	Backflow against atmospheric pressure
7	4	" "	1.0	" " " "
8	4	1.5	2.2	" " " "
9	4½	—	2.5	—
10	4½	2.0	2.5	Backflow against pressure of 1.6
11	5¾	6.5	7.5	" " " " 6.0 (severe edema)
12	19	1.0	1.5	" " atmospheric pressure
13	20	—	6.0	No backflow; severely inflamed ear
14	20	3.4	4.0	Backflow against pressure of 2.8
15	22	—	13.8	No backflow; beefy ear
16	22	—	12.5	" " " induration of ear
17	22	—	6.0	" " " " " "
18	24	—	10.0	" " " " " "
19	24	1.5	2.0	Backflow against atmospheric pressure
20	24	3.5	4.0	" " " "
21	24	2.8	3.5	" " " "
	days			
22	2	—	6.0	No backflow
23	2	—	13.5	" " ; beefy ear
24	4	—	3.0	" "
25	5	Not measurable	1.5	Backflow against atmospheric pressure
26	5	" "	1.5	" " " "
27	7	" "	1.8	" " " "
28	7	2.0	2.2	" " pressure of 1.8
29	11	Not measurable	1.6	" " atmospheric pressure
30	11	—	2.1	No backflow; edema very slight

appeared only in ears showing a distinct inflammatory reaction, as in Experiments 13 and 15 to 18, inclusive. It also appeared in one ear tested after 48 hours (Experiment 23). In instances in which the ears appeared beefy there was no backflow of freely movable edema fluid. In all these, as indicated in the table, inflammation was severe and the interstitial resistance (column 4), was high. It is noteworthy that although edema of inflammation may be intense and the interstitial resistance high, yet no free fluid may be present.

By contrast, in instances showing slight or no increase of interstitial resistance, edema was plainly perceptible and fluid flowed from the tissues into the injecting apparatus when the latter was opened to atmospheric pressure. Obviously the tissues must have stretched to accomodate the edema fluid. The phenomenon appeared in several instances of edema of short duration (Experiments 2, 4, 6, 7, 8, 12, and 19), and in most of the experiments made 4 days or more after the edema-forming injury (the last 7 experiments of Table III). In all but 2 of the latter edema fluid flowed into the injecting apparatus against atmospheric pressure, although in most instances the pressure of the edema fluid was too low to be measured accurately.

Experiments 1 to 11 (Table III) showed that the elevation of edema fluid pressure and interstitial resistance, observed in the preceding experiments (Table II), during the first hour after painting the ears with xylol and during the formation of edema, was not maintained during the next few hours. Reference to some of the findings in Table II, Experiments 7, 10, and 11, shows that in these instances the pressures had begun to fall by the 60th minute.

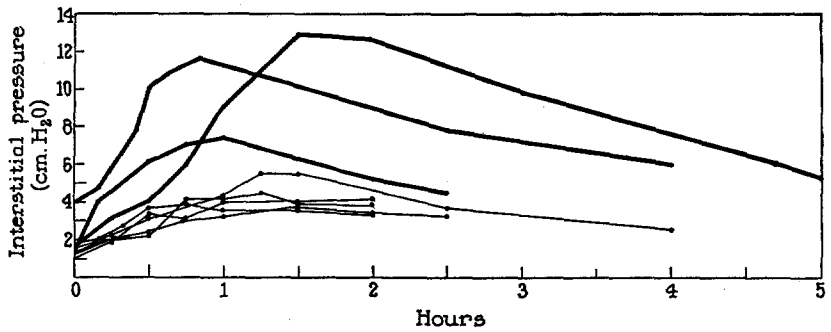
In contrast, 18 to 48 hours after injury had taken place (Experiments 12 to 23 in Table III), the interstitial resistance either rose again, if the reaction to the injury was severe and inflammatory in nature (Experiments 13 to 18, 20, 22, 23), or remained low if the reaction was mild (Experiments 12, 19, 21). Finally when resolution of the edema occurred, the interstitial resistance fell to normal levels (Experiments 26 to 30) in spite of the fact that the tissues remained edematous.

The Rise and Fall of Edema Fluid Pressure Following the Introduction of Irritating Solutions into the Skin.—Potent edema-forming fluids, to be described below, were brought at atmospheric pressure into contact with the skin of the ears of 8 mice. Then, at intervals during several hours, edema fluid pressure determinations were made, using the edema-forming solutions themselves as the test fluids in the apparatus.

For 5 of the experiments the edema-forming fluid consisted of Locke's solution containing 5.5 per cent of pontamine sky blue. As already stated, ¼ to ½ per cent of the dye in Locke's solution elicits a mild edema when introduced into the skin. For the other 3 experiments, 3 volumes of a 21.6 per cent aqueous solution of this dye were employed, mixed with 1 volume of a 10 per cent extract of bull's testicle, as prepared by Duran-Reynals (26). The latter material contains an active "spreading factor" which brings about a marked edema when injected into skin.

Text-fig. 2 summarizes the findings. The fine lines indicate the changes in the edema fluid pressure occurring in the 5 experiments in which the 5.5 per

cent dye-Locke's solution was brought into contact with the dermal tissues. The heavy lines depict the results in the 3 experiments with the mixture of dye and testicle extract. During the tests it was easy to see under the microscope that the mixture of testicle extract and concentrated dye solution induced edema far more rapidly than did the 5.5 per cent dye solution alone. The edema fluid pressures obtained with the former were regularly higher than those produced by the latter, although with both test fluids the pressures reached their highest levels in about the same period of time, that is to say at some time between 45 minutes and 1½ hours after the introduction of the irritant. Thereafter the pressures declined as the irritant became diluted with edema fluid.



TEXT-FIG. 2. Changes of edema fluid pressure in mouse skin after bringing powerful edema-forming solutions into contact with the interstitial tissue at atmospheric pressure. (See text.)

Interstitial Resistance in the Intradermal Tissue after Trauma

In 36 experiments the interstitial resistance was determined in the skin of the ears of mice at various intervals after a standardized pressure injury.

The mice were lightly anesthetized with ether. An area of the tip of one ear, 4 × 8 mm., was placed above the lower, corrugated blade of a pair of surgical forceps, lying on the table. The upper blade was brought down upon the upper surface of the ear which was then pinched between the blades for 30 seconds by lightly holding a 500 gm. weight on the tip of the upper blade. The animals were allowed to come out of ether and interstitial resistance and edema fluid pressure determinations were made at various intervals thereafter, under nembutal anesthesia. The procedure brought about an immediate hyperemia of the ear followed by edema for the first few hours. Later there occurred in some instances beefy induration and localized necrosis. In general necrosis did not appear and repair began to take place after the 2nd or 3rd day.

All interstitial resistance or edema fluid pressure measurements were made with the ½ per cent dye-Locke's solution. The findings are summarized in Table IV.

In 4 of the 36 experiments (Table IV, Experiments 5, 9, 11, and 31), the highest measurements of interstitial resistance were obtained that we have en-

TABLE IV

Intradermal Interstitial Resistance and Edema Fluid Pressure in the Traumatized Ear of the Mouse

Exp. No.	After injury	Edema fluid pressure	Interstitial resistance	Comments
		cm. of water	*cm. of water*	
1	15 min.	—	2.5	Intense hyperemia
	40 "	3.0	3.5	
2	½ hr.	5.5	6.0	Severe edema
3	1 "	8.0	8.5	" "
4	3¾ hrs.	—	3.6	In an area of hemorrhage
			6.3	In a clear, edematous area
5	3¾ "	No backflow	29.0	Very high interstitial pressure; finding confirmed in another area of the ear. Very severe injury, ear beefy, no demonstrable edema fluid. Progressed to necrosis later
			33.0	
6	4 "	8.8	9.5	Edema, not beefy
7	4⅓ "	—	15.0	Severe reaction
8	4⅓ "	11.2	12.0	In two areas of the ear
		—	14.0	
9	18 "	No backflow	36.2	No demonstrable edema fluid. Beefy induration, progressed later to necrosis
10	18 "	" "	14.5	Severe injury. No demonstrable edema fluid. No later necrosis
11	18 "	" "	46.0	No demonstrable edema fluid. Beefy induration, progressed to necrosis later
12	18 "	" "	4.7	Very little reaction to injury. No demonstrable edema fluid
13	18 "	" "	18.0	Severe reaction. No demonstrable edema fluid
14	19 "	3.2	3.6	Very little reaction
15	19 "	—	3.0	" " "
16	19 "	10.9	11.7	Severe reaction, beefy, but edema fluid present
17	21 "	—	12.1	Moderately severe reaction
18	22 "	9.8	10.0	" " "
19	24 "	No backflow	11.7	" " " . No demonstrable edema fluid
20	24 "	—	6.3	Moderately severe reaction, but low pressure
21	24 "	8.9	10.4	" " "
22	46 "	1.5	1.9	Healing
23	47 "	5.9	6.6	More severely injured than the ears of Nos. 12, 14, or 15
24	48 "	—	3.1	—
25	48 "	—	2.8	—
26	48 "	No backflow	10.0	Beefy ear. No demonstrable edema fluid. No necrosis
27	48 "	" "	12.0	" "

TABLE IV—*Concluded*

Exp. No.	After injury	Edema fluid pressure	Interstitial resistance	Comments
		cm. of water	*cm. of water*	
28	66 "	—	2.3	Healing, reaction subsiding
29	67 "	—	4.4	" " "
30	69 "	2.2	2.7	" " "
31	3½ days	No backflow	37.0	Beefy induration on way to necrosis. No demonstrable edema fluid
32	4 "	—	4.2	Reaction subsiding
33	5 "	4.7	5.2	" "
34	6 "	No backflow	11.1	Reaction subsiding but still high pressure. No demonstrable edema fluid
35	7 "	" "	4.6	Reaction subsiding. No demonstrable edema fluid
36	11 "	" "	4.5	" "

countered, 29.0 to 33.0, 36.2, 46.0, and 37.0 cm. of water respectively. Indeed in certain of these the interstitial resistance was higher than the hydrostatic pressure of blood at the arterial end of the capillary, as measured by Landis (14–16). There was no perceptible extravasation of blood in these ears nor was the skin visibly necrotic. Instead a beefy induration developed in about 18 hours and later progressed to necrosis. At the time of the induration there was no visible backflow of fluid into the injecting apparatus when the latter was opened to the room air, but the dermal tissue was swollen and exhibited a marked resistance to the entrance of fluid even in minute amounts. It is of great interest that in just these instances, in which the interstitial resistance was very high and when consequently there could be little or no escape of fluid from the blood vessels, necrosis was found later. The relationship will be investigated in the future. At present we are unable to state whether or not thrombosis of the vessels had occurred.

In the other instances in which severe reactions were noted, the interstitial resistance and edema fluid pressure were found higher than in skin in which edema of irritation had rapidly formed. Most of the tests were made $3\frac{3}{4}$ hours to 2 days after the injury showed the interstitial resistance to be well elevated, and so too was the edema fluid pressure in the instances in which there was enough free edema fluid to allow a pressure measurement to be made. In later tests carried out as the reactions subsided, the pressures fell to levels only slightly above that of the normal interstitial resistance.

Certain other instances showed a severe tissue reaction 18 hours after the injury and no demonstrable free fluid, Experiments 10 and 13 for example, yet the ears healed and did not become necrotic. In these animals, the interstitial

resistance rose only to 14.5 and 18.0 cm. of water, respectively. Another instance, Experiment 16, showed a severe reaction to the injury but free edema fluid was demonstrable. In this ear, too, no necrosis occurred and the edema fluid pressure was not very great, 10.9 of water.

The Interstitial Resistance in Living Human Skin

Several workers have investigated pressure conditions within the subcutaneous tissues of man but the pressures within the dermis itself have aroused but little interest, as has been mentioned earlier in this paper. Because of the scantiness of the recorded data it seemed well to measure the intradermal interstitial resistance by the technique here described. No measurements of subcutaneous pressures were attempted. Extensive studies were not undertaken since the method, designed primarily for use with small animals, would require special adaptations for frequent use in the clinic. Accurate determinations of the interstitial resistance take 10 to 15 minutes for the first determination and 3 to 4 minutes for each subsequent one. During this period, it is necessary for the subject to maintain complete immobility. The resulting postural discomforts are too great to be required of patients.

Changes Made in the Apparatus in Order to Determine the Interstitial Resistance in Human Skin.—The amount of fluid introduced into human skin to measure the interstitial resistance was the same as that used for the animal tests and consequently so small that it was necessary to rule out temperature effects upon the movement of the meniscus of the fluid in the pipette. As described elsewhere (1), this was done in the foregoing experiments by submerging the pipette in a constant temperature bath and allowing the tip of the needle to emerge through a water-tight seal into an open glass dish. The experimental animals, placed in the dish, were brought to the level of the needle without having to be submerged in the water. For tests on human beings the apparatus had to be modified. The water bath was discarded and, in order to rule out temperature changes in the pipette, it was enclosed in a celluloid box. A second box, 1 cm. larger in each dimension, enclosed the first leaving an air space about it, and a third box, in turn slightly larger than the second, enclosed the latter. The injecting needle emerged from the end of the triple casing and the objectives of the microscope passed through sealed openings in the tops of the boxes so that the movement of the meniscus of the fluid in the pipette could be observed. When the variations of the room temperature were held within 0.6°C. or less the temperature of the air about the pipette in the innermost box varied less than 0.2°C.

The amount of contraction or expansion of the fluid in the pipette under these circumstances was without important effect upon the estimations of interstitial resistance.

The Tests.—All tests were made with ½ per cent dye-Locke's solution. This mixture was brought into contact with the dermal tissue of 3 unanesthetized human beings in regions of the body to be stated below. As in the case of mice and rabbits, the dye-Locke's solution failed to enter the skin at atmospheric pressure, save in rare instances, but passed in continuously at low pressures. In 8 tests, the interstitial resistance was measured in the skin of the volar surface of the forearm, and in 2 tests in the skin of its dorsal surface, the forearm in each trial resting on a table at the level of the apex beat of the heart. Two interstitial resistance determinations were made in the skin over the deltoid muscle of a sitting subject, and 5 in the skin of the dorsal surface of the ankle while the subject lay on his back. Of the 17 tests

carried out, 10 were made on one subject and 3 and 4 respectively on each of two others. No marked individual differences were found.

In the forearm, intradermal interstitial resistances of 2.5, 2.9, 3.0, 3.0, 3.2, 3.7, 4.5, and 5.1 cm. of water were encountered on the volar surface, and resistances of 2.1 and 2.5 on the dorsal surface. In the upper arm, in tense skin over the deltoid muscle, we found the interstitial resistance as high as 5.0 and 6.7 cm. of water, and on the dorsum of the ankle, with the subject lying flat, it was 2.5, 2.9, 3.0, 3.1, and 3.5 cm. of water.

In normal human skin, the interstitial resistance is higher than in the skin of the mouse. Even so, the resistances found by us are significantly lower than the pressures reported by the only authors who have discussed the intradermal tissue pressure in man (8–11). The difference is to be attributed no doubt to the employment of such a minute amount of fluid in the tests here reported that appreciable artificial pressures were not set up.

DISCUSSION

The pressure existing in the normal cutaneous tissues cannot be directly measured by methods now at our disposal because of the lack of enough freely movable extravascular fluid to make manometric determinations. This difficulty has been circumvented, for practical purposes, by the employment of methods (1–3) for the introduction of almost microscopic amounts of relatively unabsorbable fluids into tissues, in such a manner that they do not enter directly into either blood vessels or lymphatics. The pressure required to overcome the resistance of the tissues to the movement of the introduced fluid at a relatively constant rate, about the slowest that could be accurately measured, has been termed the interstitial resistance. Measurements of the interstitial resistance of the skin under various conditions have shown something about the true pressures in the skin.

The Relationship of Interstitial Resistance to Interstitial Pressure.—One cannot state from our experiments how much higher the interstitial resistance is than the interstitial pressure prevailing in normal skin. Our observations upon edematous skin, in which free fluid was present, have shown in scores of instances (see Table II to IV) that the interstitial resistance of edematous mouse skin was usually about 0.5 cm. of water higher than the edema fluid pressure which is equal to the interstitial pressure. In work to be reported later, the same difference was found in human skin. It does not follow that in normal skin the relationship is the same, but one can state that the interstitial pressure must be only slightly less than the interstitial resistance; therefore in normal human skin it must be slightly less, on the average, than 3.1 cm. of water, and in the mouse slightly less than 1.7 cm. of water. One may conclude that under normal conditions and with the human subject or animal at rest the effect of interstitial pressure upon fluid exchange to and from the blood is negligible. The effect upon lymph formation will be discussed in a later paper.

Under various pathological conditions, the pressures within the skin can be ascertained with certainty, as, for example, when free edema fluid is present. Pressure conditions in edematous skin vary much, depending upon circumstances. For example, chemical irritation of the skin by xylol elicited widely different reactions from animal to animal. In some, the edema fluid pressure and interstitial resistance rose so little that the effects were negligible. In others, the skin became edematous rapidly, and in about an hour pressures were recorded which were sufficient to exercise definite partial opposition to the filtration of fluid from the blood vessels and effective aid to the formation of lymph. In general, rapid edema formation led to higher edema fluid pressure and higher interstitial resistance. On the other hand, in those instances in which edema formed slowly and also in edemas of long standing, in which the tissues become boggy, neither the edema fluid pressure nor the interstitial resistance was increased. In these tissues there must have been some readjustment of the fixed elements to permit the accumulation of fluid without an increase in the pressure of the edema fluid.

Of much interest was the occasional finding of boggy edematous skin, in appearance like ordinary edematous skin, but in which no free fluid was demonstrable. In these instances there must have been swelling or imbibition of tissue fluid by the tissue elements, or clotting of the tissue fluid, resulting in an increased tissue resistance. More will be said of this in a later paper. Often, too, if the tissues became indurated following mechanical injury, a lack of free edema fluid prohibited measurement of the interstitial pressure. In these instances, however, the interstitial resistance was so high that the escape of fluid from the blood vessels must have been prevented. As already mentioned, these were the instances in which necrosis developed, perhaps primarily from the high pressure, or perhaps because of preceding thrombosis or failure of the circulation in the injured areas.

The Relationship of Interstitial Resistance and Pressure of the Edema Fluid to the Transport of Fluid through Edematous Skin.—Intradermal injections of fluid spread more rapidly through edematous human skin than through that which is normal. In consequence it is believed by some that extravascular fluid may move through edematous skin with greater facility than ordinary. But fluids injected by hand are forced into the tissues at pressures so much higher than the interstitial resistance that the conditions are not like those imposed upon edema fluid that has formed naturally. One may well ask: Is the exchange of material between the blood and tissues more rapid in edematous skin than ordinary? The present work has shown that the interstitial resistance of edematous skin is either equal to that of normal skin or higher, that is, under conditions approaching the physiological, fluid introduced from without moves through edematous skin with as much difficulty as through normal skin, and, when the extravascular pressures are higher than normal, with greater difficulty. Earlier work has shown (27, 28) that dye solutions spread more

rapidly in skin becoming edematous than through normal skin, but they spread more slowly than usual when introduced into skin already edematous, especially if the edema is of long standing. Both sets of findings indicate that the movement of substances through skin which has become edematous is not as rapid as through normal skin under physiological conditions, and the fluid exchange is probably not as efficient.

On the other hand the clinical observation that fluid injected into edematous skin spreads more rapidly than in normal skin is borne out by a consideration of some observations reported here, as also by some earlier ones. In the earlier work (3) relatively unabsorbable test fluids were forced into normal mouse skin at low pressures of 1.5 to 2.5 cm. of water and then slight increases in pressure were brought to bear. No significant increase in the rate of inflow resulted until, as mentioned above, the "breaking point" was reached at pressures of about 8.5 cm. of water and there suddenly occurred a great inflow, as though the tissues had been broken apart (3). In the present work, during experiments upon edematous skin like those summarized in Tables II and III, we forced the test fluids into the skin at gradually increasing pressures, after the interstitial resistance had been determined. In these experiments, each slight increase in pressure above the interstitial resistance, required to initiate flow, led to a significant increase of inflow, like that observed in normal skin only after the "breaking point" had been reached. In all, the rate of inflow was greater than that which took place into normal skin at similar pressures. From this it is clear that fluids introduced into edematous skin and then subjected to pressures higher than the interstitial resistance, do move through it more readily than fluids introduced into normal skin under the same circumstances.

Findings of this sort explain the experience of Meyer and Holland (9, 10) who reported a decrease of the tissue pressure in edematous skin. As indicated earlier in the present paper, these authors introduced excessive quantities of fluid into the skin at pressures not only above the interstitial resistance but even above the "breaking point." Having disrupted the boggy tissues, they found thereafter that the fluids injected by them, still at pressures above the interstitial resistance, moved more easily through the edematous skin than through normal skin; a finding which they took to indicate a lessened tissue pressure, as was not the actual case.

SUMMARY

Means have been described for the study of pressure conditions in normal and pathological skin of living human beings and mice. The true pressure in normal skin cannot be measured directly by any of the means hitherto described, because there is insufficient free fluid to make manometric determinations. However, for practical purposes, the intracutaneous pressure has been approxi-

mately estimated by introducing into skin exceedingly small amounts of a relatively unabsorbable fluid, a mixture of Locke's solution and a vital dye, and then finding the least pressure required to overcome the resistance of the skin to the passage of this fluid through it at the lowest rate measurable with accuracy by the apparatus at hand. In the present paper measurements of this pressure have been termed the interstitial resistance.

In normal skin the interstitial pressure, as estimated by measurements of the interstitial resistance, is low, slightly less, on the average, than 1.7 cm. of water in the skin of the mouse, and less than 3.1 cm. of water in human skin. It remains unchanged in states of active hyperemia.

In edematous skin the interstitial pressure can be directly measured by determination of the edema fluid pressure. It has been compared with determinations of the interstitial resistance and found to be only 0.5 cm. of water lower in both the mouse and man.

Under the conditions of our experiments, in skin rendered slowly edematous by the introduction of irritant chemicals or their topical application, little rise in pressure took place. On the other hand, in rapidly forming edema of the skin the edema fluid pressure and the intradermal interstitial resistance rose and became great enough to hinder materially the further escape of fluid from the blood vessels. The edema fluid pressure rose in proportion to the rapidity with which the edema formed. When a rapidly formed edema subsided, the edema fluid pressure and interstitial resistance fell, but if inflammation and induration followed later, the interstitial resistance became high again. As these conditions subsided the interstitial resistance fell, at times to normal levels, even in the presence of edema.

In mouse skin injured by squeezing according to a standard procedure, with result in pronounced edema, the intradermal interstitial resistance rose within a few hours to levels of 10 to 15 cm. of water. In those instances in which the injury progressed to induration, the interstitial resistance rose to such high levels that it seemed impossible that fluid could continue to escape from the capillaries. Such a state of affairs may be of great importance in determining whether necrosis follows trauma.

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