THE MOVEMENT OF ELECTROLYTES AND OF WATER IN
SURVIVING TISSUE OF THE LIVER

BY JOHN D. BROOME, M.B., AND EUGENE L. OPIE, M.D.

(From The Rockefeller Institute)

(Received for publication, May 12, 1960)

Liver tissue and kidney cortex when immersed in solutions of sodium chloride
immediately after their removal from the body have been found to be isotonic
with solutions that have approximately twice the concentration of the sodium
chloride in physiological salt solution or in blood plasma (1, 2) but the condi-
tions that determine this relation have received widely varied explanations.

Most of those who have studied the water exchange of tissue slices immersed
in various media have attached little significance to the progress of the changes
which occur in tissues immediately after their removal from the body. Liver im-
mersed in an approximately 0.34 molar solution of sodium chloride remains in
water equilibrium with the medium during 15 to 20 minutes and then gradually
increasing water intake (1, 2) follows. When slices are immersed in a Krebs-
Ringer (3) solution made isotonic with liver by addition of sodium chloride, the
period of water equilibrium may be prolonged to 1 or 2 hours because, as it ap-
ppears, the tissue has undergone less injury in this medium than in a solution of
sodium chloride (3) with the same molar concentration.

Rapid weighing of slices of liver or of kidney of the white rat in tenths of a
milligram by means of a torsion balance facilitates the measurement in per cent
of loss or gain of water between tissue and the immersing medium. Methods
used in the present experiments for the study of water movement have been
described in detail (1, 2). If slices are weighed with appropriate precautions be-
fore and after immersion, their solid content is measurable by drying and con-
irms the belief that changes in the per cent of total weight are referable to gain
or loss of water.

Most of those who have measured the dry weight of tissues have dried tissues
overnight or longer at a temperature of approximately 105°C. Following im-
mersion in Krebs-Ringer solution the dry weight of tissue may undergo consid-
erable diminution. Loss of total nitrogen by slices of tissue immersed in media
applicable to the supravital manometric determination of oxygen consumption
has been measured by Cutting and McCance (4), by Robinson (5), and by
Aebi (6) and confirms the loss of solid substances that is indicated by changes
in dry weights.
Movement of Electrolytes between Liver Slices and Sodium Chloride Solutions

Slices of liver have been immersed during periods of 10 minutes in solutions of sodium chloride varying from 0.1 to 0.5 molar, in order to determine the concentration that is in water equilibrium with the tissue. The flame photometer has been used to measure the movement of sodium and potassium ions between the tissue and the medium.

![Graph](image)

**Fig. 1.** Experiment 1.—Per cent of weight (on left of graph) measuring water exchange is indicated by a heavy continuous line which crosses the base line at the point of water equilibrium. Changes in the tissue content of sodium (with scale at right) is indicated by a broken line in m.eq./kg., and of potassium by a dotted line. The column at the left of the graph shows the relation of sodium and of potassium to the tissue weight before immersion.

Tissue slices, weighing approximately 200 mg. after removal of surplus fluid by contact with cotton gauze, were placed in stoppered bottles containing 3 ml. N/10 sulfuric acid. After standing a minimum time of 24 hours, the bottles were thoroughly shaken and 0.5 ml. portions of the supernatant were removed for dilution with water to a degree suitable for estimation of sodium or potassium in the flame photometer. A Coleman model 21 flame photometer was used.

Samples (0.5 ml.) of the supernatant described above were analyzed for chloride ion by the silver iodate method of Sendroy as modified by Hiller and Van Slyke (7).
Selected experiments will be described and designated by numbers assigned with no relation to the order in which they were performed.

The liver tissue in Experiment 1 (see Fig. 1) is isotonic with 0.3 molar sodium chloride. Slices have contained 23.6 m.eq./kg. of sodium and 94.1 m.eq./kg. of potassium. When they have been immersed in distilled water, fluid has entered rapidly and there has been loss of both ions. In graded solutions of sodium chloride water intake of the tissues has diminished with increasing concentration while sodium has increased continuously. In solutions hypertonic for the tissue, sodium has entered the tissue in quantity increasing with each tested concentration and water intake has diminished.

| TABLE I |

Water Exchange and Movement of Electrolytes in Liver Slices of Experiment 2 Immersed in Water and in Graded Solutions of Sodium Chloride during 10 Minutes

<table>
<thead>
<tr>
<th>Molar concentrations of solutions of sodium chloride</th>
<th>0.15</th>
<th>0.2</th>
<th>0.3</th>
<th>0.4</th>
<th>0.5</th>
</tr>
</thead>
<tbody>
<tr>
<td>Per cent of initial weight</td>
<td>100.0</td>
<td>114.2</td>
<td>113.0</td>
<td>104.3</td>
<td>98.0</td>
</tr>
<tr>
<td>Dry weight in per cent of slice</td>
<td>32.6</td>
<td>25.0</td>
<td>25.3</td>
<td>28.7</td>
<td>31.8</td>
</tr>
<tr>
<td>m.eq./kg. sodium</td>
<td>31.8</td>
<td>66.5</td>
<td>76.0</td>
<td>122.5</td>
<td>157.3</td>
</tr>
<tr>
<td>m.eq./kg. sodium in tissue water</td>
<td>47.2</td>
<td>88.7</td>
<td>101.7</td>
<td>171.7</td>
<td>230.6</td>
</tr>
<tr>
<td>m.eq./kg. potassium</td>
<td>95.0</td>
<td>53.0</td>
<td>72.5</td>
<td>64.8</td>
<td>72.5</td>
</tr>
<tr>
<td>m.eq./kg. potassium in tissue water</td>
<td>140.9</td>
<td>69.3</td>
<td>97.1</td>
<td>91.1</td>
<td>106.3</td>
</tr>
</tbody>
</table>

The original potassium content of the liver slices in this experiment has been 94.1 m.eq./kg. It has diminished after 10 minutes of immersion in 0.1 molar sodium chloride solution to 52.1 m.eq./kg. In graded solutions of sodium chloride with increasing concentration, potassium ions have maintained a nearly even level whereas under the same conditions water intake has increased when the medium has been hypotonic and diminished when hypertonic.

In Experiment 2 (Table I, Fig. 2) liver slices have been found to be isotonic with 0.36 molar sodium chloride by the procedure which has been described. Initial sodium in the liver tissue has been 31.8 m.eq./kg. and in the tissue water 47.2 m.eq./kg. When slices have been immersed in distilled water for 10 minutes sodium has diminished slightly and water has entered rapidly. After immersion in graded solutions of sodium chloride the sodium content of the tissue has increased continuously with the concentration of the medium but water intake in the hypotonic solutions (0.1 and 0.2 M) has diminished whereas in hypertonic solutions (0.4 and 0.5 M) sodium intake has increased along with loss of water.
In sodium chloride solutions with increasing concentration, potassium has maintained an approximately even level whereas water intake has diminished.

Movement of Water and of Electrolytes in Liver Slices Immersed in Krebs-Ringer Solution

The procedure used in the following experiments (Nos. 3 to 5) has been described in detail elsewhere (3) and will be cited briefly. When liver slices from the rat have been immersed in Krebs-Ringer solution in which electrolytes have been adjusted to the molar concentration of 0.15 molar with phosphate buffer

![Graph showing changes in weight percentage and electrolyte content over molar concentration.]

Fig. 2. Experiment 2.—Changes in per cent of weight measuring water and in content of sodium and of potassium are shown as in Fig. 1. Sodium and potassium ions in the tissue and in tissue water are indicated by broken and by dotted lines. The column at the left of the graph shows their quantity in the tissue and in the tissue water before immersion.

and oxygen 100 per cent allowed to bubble through the medium, entrance of water indicated by increased per cent of weight proceeds rapidly and with some variation reaches a maximum after about 2 hours. Slight diminution occurs after 3 hours and still more after 4.

The dry weight of the tissue slices determined by heating to a constant weight has been used to measure the relation of fluid to solid contents of the tissue after various periods of immersion. In view of changes in water content of the slices the dry weight of slices after immersion is not a measure of subsequent changes in the solid content of the slices. The initial dry weight of the slices may be used as a standard for comparison.

In Experiment 3 (Table II, Fig. 3) liver slices have been immersed in Krebs-
Ringer solution, 0.154 molar, hypotonic in relation to the liver tissue and water has entered the tissue. Sodium and chloride ions, which have measured in the liver 25 and 30 m.eq./kg, respectively, have passed rapidly, with approximately parallel increase, from the medium into the tissue and after 1 hour it has con-

![Graph](image)

FIG. 3. Experiment 3.—Changes in water movement are shown by a continuous line, in sodium ions by a broken line, in potassium ions by a dotted line, and in chloride ions by a line broken and dotted.

### TABLE II

Movement of Water and Electrolytes in Liver Slices of Experiment 3 Immersed in Krebs-Ringer Solution 0.154 M with Phosphate Buffer and Oxygen

<table>
<thead>
<tr>
<th>Per cent of weight</th>
<th>Control</th>
<th>Minutes of immersion</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>100</td>
<td>132</td>
</tr>
<tr>
<td>Dry weight in per cent of slice</td>
<td>32.85</td>
<td>29.25</td>
</tr>
<tr>
<td>m.eq./kg. sodium in slices</td>
<td>25</td>
<td>100</td>
</tr>
<tr>
<td>m.eq./kg. sodium in tissue water</td>
<td>37.2</td>
<td>141.3</td>
</tr>
<tr>
<td>m.eq./kg. potassium in slices</td>
<td>96</td>
<td>40</td>
</tr>
<tr>
<td>m.eq./kg. potassium in tissue water</td>
<td>142.9</td>
<td>56.5</td>
</tr>
<tr>
<td>m.eq./kg. chloride in slices</td>
<td>30</td>
<td>80</td>
</tr>
<tr>
<td>m.eq./kg. chloride in tissue water</td>
<td>44.7</td>
<td>113.1</td>
</tr>
<tr>
<td>m.eq./kg. sodium and potassium in tissue water</td>
<td>180.1</td>
<td>197.8</td>
</tr>
</tbody>
</table>
tained sodium ions 100 m.eq./kg. and chloride 80 m.eq./kg. At the same time the sodium and chloride concentrations in the tissue water of the slice approximate those in the immersion fluid and have been 141.3 and 113.1 m.eq./kg. respectively. During the next hour sodium and chloride have remained little changed, but after 180 minutes they are increasing at a time when water con-

![Graph](image)

**Fig. 4.** Experiment 4.—Liver slices have been immersed in Krebs-Ringer solution with isotonicity increased to 0.35 molar by addition of sodium chloride. Movement of water and of sodium, potassium, and chloride ions are shown as in Fig. 3.

tent is rapidly diminishing. Potassium ions, which have been 96 m.eq./kg. in the tissue before immersion, are during 1 hour rapidly lost, but after 2 and 3 hours remain nearly constant, that is, from 24.9 to 20.8 m.eq./kg. or in the tissue water 33.3 and 34.1 m.eq./kg. During the period of the experiment the measured dry weight of the tissue has diminished with the increased water content of the slices. When this is compared with the initial dry weight of the tissue its solid content as determined by drying is found to have decreased about one-third
(Table II). Sodium and chloride ions follow a parallel course with no evident relation to that of potassium.

In the experiments which follow, the movements of water and of sodium, potassium, and chloride ions have been measured in liver slices, immersed in media made approximately isotonic with them by addition of sodium chloride. In Experiment 4 (Fig. 4) with the Krebs-Ringer solution adjusted to 0.35 molar, isotonicity of the tissue has been maintained during 1 hour. At this
time when the tissue is taking in no water, sodium and chloride ions are entering rapidly, and in close approximation. Potassium ions diminish during the first 2 hours of immersion and reach a level only slightly less than that attained when liver slices have been immersed in Krebs-Ringer solutions with the isotonicity of blood plasma (Experiment 3). After 3 hours of immersion, tissue water has a potassium concentration of 25.4 m.eq./kg, that of the medium being approximately 5 m.eq./kg.

In a similar experiment, liver slices all from the same animal, Experiment 5 (Fig. 5), have been immersed, one part in Krebs-Ringer solutions with concentration increased by addition of sodium chloride to 0.3 molar, and another part in a similar solution, 0.35 molar. Immediately following immersion the 0.3 molar solution is found to be hypotonic and the 0.35 molar solution nearly isotonic with the tissue. In the weaker medium during the first 10 minutes of immersion the tissue has taken in a small quantity of water. At this time sodium and chloride have entered very rapidly but after 10 minutes more slowly. This change in the rate of entrance of the two ions has occurred when the sodium content of the tissue has reached about 152 m.eq./kg. the corresponding quantity in the tissue fluid being 212.3 m.eq./kg.; similar figures for chloride are 118 and 167.6 m.eq./kg. In the stronger solution (0.35 m) a small quantity of water has been lost during the first 10 minutes of immersion. Within 5 minutes sodium of the tissue has increased to 155 m.eq./kg. and chloride to 122.8 or in tissue water to 215.6 and 170.8 m.eq./kg. respectively. During the next 5 minutes these levels have remained almost unchanged and have been followed by rapid increase of sodium and of chloride ions.

It is noteworthy that water intake after the first 10 minutes has been greater in the medium of lower molar concentration and intake of sodium and of chloride have been less. Movement of potassium has been similar in the two solutions but in the more concentrated, like that of sodium and of chloride, it has remained unchanged during the period from 5 to 10 minutes.

Movement of Water and of Electrolytes in Liver Slices Immersed in Media with Varied Potassium Content

Liver tissue and kidney cortex have been found to be isotonic with a solution of potassium chloride of slightly greater molar concentration than that of sodium chloride isotonic with the same tissues (2). Liver has been in water equilibrium with a potassium chloride solution 0.35 molar and kidney with 0.3 molar, the corresponding figures for sodium chloride being 0.34 and 0.23 molar.

Experiments have been undertaken to determine the effect of diminished or increased quantities of potassium on water exchange and on movement of electrolytes in liver slices immersed in solutions otherwise resembling the Krebs-Ringer medium.

A solution with potassium reduced to minimal traces (Experiments 6 and 7)
has been obtained by replacing potassium chloride of the Krebs-Ringer solution
with sodium chloride of the same molar strength and substituting for potas-
sium monobasic phosphate the similar sodium salt. The movement of water and
of electrolytes has been measured in liver slices immersed in Krebs-Ringer
solutions with potassium reduced to minimal traces or increased 10-fold, and in
both instances compared with liver tissue in the usual Krebs-Ringer solution.
In each of the experiments that will be described, all determinations have been
made with liver from the same animal.

![Graphs showing water and electrolyte movement](image)

**Fig. 6.** Experiment 8.—The graph on the left shows the per cent increase of water and
m.eq./kg. of sodium, potassium, and chloride ions in liver slices after immersion in Krebs-
Ringer solution with the usual potassium content. That on the right shows changes after
immersion in Krebs-Ringer solution with potassium increased 10-fold. Per cent of weight of
slices and sodium potassium and chloride are indicated as in Fig. 1.

In Experiments 6 and 7 water exchange, intake of sodium, and loss of potas-
sium during 3 hours, have been almost the same in the medium with potassium
approximately eliminated and in the usual Krebs-Ringer solution. In the
solution with 10-fold potassium chloride, water movement has not differed
decisively from that in the other two solutions. Movement of water, sodium,
and potassium has taken almost the same course in the two experiments during
the 1st hour of immersion. In both there has been scant difference in media
with potassium reduced to a minimum and with Krebs-Ringer solution of the
usual content. With longer immersion the rate of intake of water, potassium,
and sodium changes abruptly and, though the intake of sodium and loss of potassium continue to increase, the water content of the tissue maintains an approximate level or diminishes. In both experiments in Krebs-Ringer solution with 10-fold potassium chloride, liver takes in less sodium and loses less potassium than in the two media with less potassium chloride. Nevertheless, loss of potassium has not been prevented. Though sodium intake has been less and potassium loss less in the medium with 10-fold potassium chloride, as compared with that in the other two immersion fluids, water intake has shown no evident change.

In Experiment 8 (Fig. 6) movement of water and of sodium, potassium, and chloride ions in liver slices immersed in Krebs-Ringer solution with 10-fold content of potassium chloride have been compared with corresponding changes in slices immersed in a Krebs-Ringer solution containing the usual quantity of potassium chloride. In the solution with the greater potassium content, liver has lost less potassium and less sodium has entered so that the sum of the two ions is greater after immersion in the solution with greater sodium chloride concentration. In this experiment, retention of potassium by liver slices in the medium with increased potassium chloride concentration has been accompanied by considerably less water intake than that of slices in the usual Krebs-Ringer solution. It is noteworthy that the quantity of chloride ions which enter the tissue from the unchanged Krebs-Ringer solution as in preceding experiments is

---

**Fig. 7.** Experiment 9.—Per cent of weight and m.eq./kg. of sodium, potassium, and chloride indicated as in Fig. 3.
less than the quantity of sodium ions, and the two follow an approximately parallel course. In liver after immersion in the solution with 10-fold increase of potassium chloride, chloride ions accumulate in relatively greater quantity than sodium ions and presumably have been associated with both sodium and potassium.

_Liver Tissue of Rat in Krebs-Ringer Solution at 38° and at 0°C._

When slices of the liver of four rats have been immersed in Krebs-Ringer solution at 38°C, the average water intake has increased during 2 hours and

![Graph](image)

Fig. 8. Experiment 10.—Per cent of weight and m.eq./kg. of sodium, potassium, and chloride indicated as in Fig. 3.

has reached an approximate level, maintained for another hour. After 3 hours the tissue has lost water. Under similar conditions at 0°C, water intake of liver slices from the same animals has proceeded more rapidly and after 3 hours has reached a much higher level.

In Experiment 9 (Fig. 7) liver slices have been immersed in Krebs-Ringer solution with bicarbonate buffers and gassed with 95 per cent oxygen and 5 per cent carbon dioxide allowed to bubble into a flask containing 25 cc. of the medium. During the 1st hour of immersion there has been little difference between water intake at 38° and at 0°C. During the same period sodium has entered the tissue with great rapidity and potassium has been lost at about the same rate, but gain of sodium and loss of potassium have been greater at 38°C.
than at 0°C. After 1 hour a conspicuous change occurs in the movements of water and of sodium, the rate of intake of sodium diminishing abruptly. Loss of water at 38°C is coincident with abundant intake of sodium and maximum loss of potassium, whereas continued intake of water at 0°C occurs with less intake of sodium and less loss of potassium.

In Experiment 10 (Fig. 8) with conditions similar to those of Experiment 9, water intake has differed little during the 1st 2 hours of immersion at 38°C and 0°C. In both experiments increased water intake at 0°C is associated with less entrance of sodium and greater entrance of potassium than at 38°C.

![Graph](image)

**Fig. 9.** Experiment 11.—Per cent of weight and changes in sodium, potassium, and chloride (as indicated in Fig. 1) in liver slices of rabbit immersed in Krebs-Ringer solution at 38°C and 0°C.

**Effect of Temperature on Water Movement of Liver Tissue of Rabbit Immersed in Krebs-Ringer Solution**

When liver slices in Experiment 11 (Fig. 9) from the rabbit have been immersed in Krebs-Ringer solution, 0.154 molar, water intake has been considerably greater with temperature maintained at 0°C than in the same solution at 38°C. On the contrary, sodium has entered the tissue in greater quantity at 38°C than at 0°C, and it is evident that there has been no correlation between water intake and increase of sodium. Much greater intake of water at 0°C than at 38°C has been associated with a slightly greater loss of potassium.
DISCUSSION

More than a century ago Liebig (8) found abundant potassium in muscle, but much more sodium than potassium in the blood. Gamble and his associates (9) compared the fixed bases of human serum with those recorded by Katz (10) in human striated muscle and found that the total ionic concentration of sodium, potassium, calcium, and magnesium in the water content of muscle was almost the same as that in blood serum. The water of the muscle contains about 150 m.eq. of potassium per kg. of water, 40 of magnesium, and not more than 10 of sodium. Organic phosphates approximate 150 m.eq./kg. Almost all of the sodium and apparently all of the chloride obtainable from striated muscle are in the extracellular space which by calculation based upon the demonstrable quantity of chloride in muscle (Hastings, 11) represents about 17 per cent of the tissue. Histological measurement of this space in tissue sections is in agreement with this estimate (Truax, 12).

The concentration of osmotically active constituents of the extracellular fluid has been found to be the same as that of plasma or of an ultrafiltrate of plasma (Hastings, 11) and a direct quantitative analysis of fluid obtained by means of a micropipette has confirmed this opinion (Maurer, 13). Osmotic pressure demonstrable within liver tissue in excess of that of blood plasma is evidently referable to the cells of the organ.

Robinson studied water intake of kidney (14) and of liver (15) tissue in vitro. In agreement with almost all who have recently studied the subject, he confirmed the evidence that liver or kidney tissue immediately after its removal from the body was in water equilibrium with solutions that had approximately twice the molar concentration of physiological salt solution or of blood plasma. By measurement of oxygen consumption with Barcroft manometers he found an inverse relation between water intake and oxygen consumption. Inhibition of oxygen consumption by potassium cyanide added to the immersion medium was accompanied by increased water intake. With addition of 2,4-dinitrophenol (16) swelling was as great as that with cyanide, though oxygen intake was increased. Renal tissue of the rat took up water from a Krebs-Ringer solution exposed to air, but lost it in an oxygenated medium at 38°C. In a later study Robinson (17) described varied conditions under which this reversal occurred.

Aebi (18) using Warburg manometers did not find a close parallel between oxygen consumption and water intake, but swelling varied with the potassium content of slices. With anoxia, in a calcium-free medium or in a medium containing less than 10 m.eq./kg. of potassium, the usual potassium gradient between cells and extracellular fluid was not maintained, loss of potassium being accompanied by swelling of the tissue.

Mudge (19) leached slices of kidney cortex during 2 to 3 hours in 0.15 molar sodium chloride aerated by a stream of air. The potassium content of the kidney
cortex which was 76 m.eq./kg, fell to a level not less than 25 m.eq./kg. Incubation of the slices in Warburg flasks at 25°C. with buffered medium containing 10 m.eq./kg. of potassium and 133 m.eq./kg. sodium was followed by reaccumulation of potassium. This reaccumulation he regarded as an example of active transport modified by oxygen uptake but not directly related to metabolic activity of the tissue.

Whittam and Davies (20), and Bartley, Davies, and Krebs (21) described the active transport of potassium and of sodium by kidney cortex of guinea pig. Slices of cortex lost potassium and took in water, but loss of potassium and increase of sodium might be reversed by conditions favorable to the metabolism of the tissue. The addition of α-keto glutarate to the medium brought about this reversal. Exchanges in many instances, the authors suggested, depend upon energy driven "pumps."

Deyrup (22) observed reversal of water intake by kidney slices when first immersed with no oxygen supply and later oxygenated in a Warburg apparatus. After 4 hours of partial anoxia at 38°C. reversal no longer occurred. Evidence was described suggesting that intracellular hypertonicity is improbable; the ingress of water accompanying solutes of the immersion medium was regarded as more likely (23).

Leaf (24) found increase of sodium in vitro occurring under conditions which impair cellular metabolism. Sodium, he found, entered the cell, exchanged on an ionic basis with potassium, and was accompanied by increase of chloride. The temporary rise in osmotic pressure within the cell manifested itself as an increase in tissue hydration. Leaf suggested that in life active extrusion of sodium from living cells maintains a higher extracellular than intracellular concentration of ions and this concentration counterbalances the osmotic effect of intracellular non-diffusible colloid.

Itoh and Schwartz (25) agreed with the opinion that cells adjust to osmotic changes with electrolyte shifts and do not react as simple osmometers.

Liver slices from rats, Riecker and his associates (26) found, were isotonic with solutions with molarity almost twice that of blood serum. In solutions of the same molar concentration swelling was slightly less at higher (36°–37°C.) than at lower temperatures (1°–2°C.). In animals dehydrated by withdrawal of drinking water during 2 to 6 days, the level of isotonicity of the liver rose and varied from 0.3 to 0.6 molar. When movement of sodium and potassium in liver slices immersed in solutions of high or low molar concentration was measured by means of the flame photometer, movement of electrolytes and swelling of cells were found to occur independently.

Parenchymatous cells of the liver and kidney immediately after their removal from the body were found (1) to act as osmometers capable of maintaining for a time isotonicity with solutions of sodium chloride with molar concentration approximately twice that of blood plasma. The molar concentra-
tion of a wide variety of electrolytes, as examples potassium, ammonium, magnesium, and lanthanum chlorides that are isotonic with liver or with kidney, is determined by their molecular weight, valency, and ion dissociation in accordance with well known conditions of osmosis (27). The plasma membrane of the cells of the organs are imperfectly semipermeable to electrolytes and in surviving cells the retarded entry of electrolytes brings about injury which is accompanied by changes in the permeability of the membrane.

RECAPITULATION

Liver slices immersed in graded sodium chloride solutions immediately after removal from the body (Experiments 1 and 2) have taken up water with no constant relation to the movement of electrolytes. In hypotonic solutions, corresponding with increase of the sodium chloride concentration of the medium, more sodium enters the tissue and water intake diminishes. In hypertonic solutions, with further increase of sodium intake the tissue loses water. When liver slices are immersed in sodium chloride solutions potassium is withdrawn from the tissue and in solutions of sodium chloride graded from 0.1 to 0.5 molar potassium remains almost the same in each solution although water content varies widely.

When liver slices have been immersed in Krebs-Ringer solution (Experiment 3) with electrolytes in the usual concentration (0.154 molar), sodium has entered the tissue until the sum of sodium and potassium ions of the tissue water are after about 1 hour approximately equivalent to those of the medium. They have remained unchanged for a time and then increased, though water content has decreased. Chloride ions follow the same course at a slightly lower level in part perhaps associated with cations present in the tissue. Under the same conditions potassium ions have been decreasing during 1 hour and later have remained almost unchanged though the water of the tissue has undergone wide variation. It is probable that the potassium remaining in the tissue is fixed in non-ionic form.

When liver slices have been immersed in Krebs-Ringer solution made approximately isotonic (Experiment 4) with liver slices by addition of sodium chloride (0.35 molar), no water has entered the tissue during 1 hour at a time when sodium and chloride have been entering rapidly and have reached a concentration approximately equal to that in the medium. In this experiment sodium and chloride ions have followed an almost identical course; potassium ions have decreased and after 2 hours of immersion maintained an even course.

Liver slices from the same animal (Experiment 5) have been immersed in part in a solution approximately isotonic with the tissue (0.35 molar) and in part in one slightly hypotonic (0.3 molar). In both instances sodium and chloride ions have entered the tissue very rapidly during the first 5 minutes of immersion, presumably because they have replaced the contents of blood
vessels and the extracellular fluid of the tissue. Their rate of entrance has changed abruptly after 5 minutes of immersion. In the more concentrated medium during the period from 5 to 10 minutes almost no sodium or chloride has entered the tissue. Later, though sodium and chloride have come in rapidly, there has been scant intake of water. In the Krebs-Ringer medium with electrolyte concentration slightly hypotonic for the liver tissue (0.3 molar) sodium and chloride ions have entered the tissue rapidly but their entrance has been retarded after another 10 minutes. From the more concentrated medium the tissue has taken up more sodium and chloride ions and less water than slices immersed in the hypotonic medium. Movement of potassium ions in the two media has been the reverse of that of the sodium ions, with similar changes of rate between 5 and 10 minutes of immersion.

Water intake from a solution with minimal potassium content (Experiments 6 and 7) has been almost the same as that from the usual Krebs-Ringer solution and there has been no significant change when potassium chloride of the medium has been increased 10 times. Movement of sodium and of potassium ions has differed very little in potassium-free and in the usual Krebs-Ringer medium. When the medium has contained 10-fold potassium chloride (Experiments 6 and 7) less potassium has been lost and less sodium has entered the tissue though water intake has not differed significantly. Decreased loss of potassium has not brought about diminished water intake. It is probable that increase of nitrogenous products of metabolism within the tissue or loss of them may modify osmotic pressure within the cells.

Comparison of graphs of Experiment 8 (Fig. 6) showing movement of sodium, potassium, chloride, and water in the usual Krebs-Ringer solution on the one hand and in that with 10-fold increase of potassium chloride on the other shows an excessive accumulation of chloride ions which have evidently entered with potassium. Decreased loss of potassium has not been accompanied by diminished water intake. It is probable that increase of nitrogenous products of metabolism within the tissue or loss of them may modify osmotic pressure within the cells.

Water intake under the conditions of these experiments has been almost the same during 1 hour of immersion when slices have been immersed at 38°C and 0°C. but, later, water has entered more rapidly at 0°C. At 38°C. increase of sodium and loss of potassium (Experiments 9 and 10) have been greater in association with less water intake.

SUMMARY AND CONCLUSIONS

When liver slices immediately after their removal from the body are immersed in graded solutions of sodium chloride, movement of water does not follow a course determined by movement of sodium ions. From hypotonic solutions sodium enters slowly and swelling proceeds rapidly but with increasing concentration entrance of sodium increases and swelling diminishes in accord with the osmotic relations between tissue and the medium.
The extracellular fluid of liver has the same osmotic pressure as blood plasma, and entrance of water into liver slices from media with greater molar concentration is determined by the intracellular pressure of the parenchymatous cells of the tissue.

The plasma membrane of the liver cell is semipermeable to electrolytes but its semipermeability is imperfect, may be impaired, and when in media isotonic with the cells some of the electrolyte enters them. With continued entrance permeability to both electrolyte and water increases and in case of sodium become evident after 15 or 20 minutes.

A medium more favorable to the tissue prolongs the period of isotonicity. In solutions with electrolytes otherwise similar to those of the blood plasma, e.g. Krebs-Ringer solution, but with molar concentration of electrolytes approximately doubled by addition of sodium chloride isotonicity may be prolonged during a period of 1 hour or more.

When potassium chloride is added to the Krebs-Ringer solution so that its potassium content has been increased 10-fold the water intake of liver cells has not varied in accord with the potassium content of the medium.

In a medium with the electrolyte contents of blood plasma (Krebs-Ringer solution) liver cells after 1 hour gain sodium and lose potassium, but later potassium maintains a nearly constant level though swelling increases.

Less sodium enters and less potassium is lost from liver cells at 0°C. than at 38°C. and 0°C. swelling is greater.

Movement of water between cells and extracellular fluid may occur independently of changes in the sodium or of potassium content of cells and doubtless is in part determined by substances associated with metabolism.

BIBLIOGRAPHY


