THE EFFECT OF SALT CONCENTRATION OF THE MEDIUM
ON THE RATE OF OSMOSIS OF WATER THROUGH
THE MEMBRANE OF LIVING CELLS.

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(Accepted for publication, February 17, 1927.)

The present paper reports a further investigation of the kinetics of osmosis, using the unfertilized egg of the sea urchin Arbacia punctulata as an osmometer. In a previous communication dealing chiefly with the affect of temperature on osmosis, it was pointed out that the rate of osmosis is influenced by a second external factor, the salt concentration of the medium (1). It is with this factor that the present investigation is concerned. The technic used was the same as that previously described.

In order to understand the mechanism involved it was desirable to study the effect of salt concentration of the medium over a wide range—from 20 per cent to 125 per cent of sea water. Hence it was obviously necessary that for low concentrations endosmosis should be studied, and for high concentrations exosmosis. This procedure was justified since both processes follow the same diffusion equation, and since their velocity constants are practically identical when the concentration of the medium is the same in the two cases (2).

Experiments.

One preliminary question needed to be answered. Suppose different lots of eggs (from the same animal) were placed in 60, 70, 80 and 100 per cent sea water

\[ 2 \frac{d\alpha}{dt} = k (a - x) \]

where \( a \) is the total volume of water that will cross the membrane before equilibrium is established, \( x \) the amount that has already crossed at time \( t \), and \( k \) is the velocity constant.

1 Sea water evaporated so that the concentration of salts is 5/4 that in 100 per cent sea water; the original pH of this solution was restored with CO₂.

The Journal of General Physiology
FIG. 1. Different lots of cells were placed in 60, 70 and 80 per cent sea water and allowed to reach osmotic equilibrium; then each lot was transferred to 40 per cent sea water, in which further swelling occurred. In each case the observational points from the several experiments fall along a single curve calculated from the equation $kt = \log \frac{a}{x}$. (In this equation the value of $a$ was calculated as previously described (1); the value of $k = 0.027$ was determined by transferring cells from full strength—100 per cent—sea water to 40 per cent sea water.)

The points of intersection of the several base lines with the curve were found by observing the mean volume at equilibrium of twenty cells in 80, 70 and 60 per cent sea water, respectively; these values correspond to volumes at time zero in the several experiments.

The values of $k$ in the several experiments were practically the same as the value of $k$ for the curve. (Each observational point represents the mean of six cells.)
and allowed to reach osmotic equilibrium, and then each lot was transferred to a
more hypotonic solution, e.g. 40 per cent sea water, in which further swelling would
occur, would the values of the velocity constants be the same in all lots,—i.e. would
volume increase go half way to equilibrium in the same length of time? Several
such experiments were made, and an affirmative answer was obtained. Thus the
values of \( k \) for swelling, on transferring the cells from the several solutions into 40
per cent sea water at 21°C., were as follows: from 100 per cent, 0.027; from 80

![Graph showing the effect of osmotic pressure of the medium on the rate of osmosis of water through the membrane of living unfertilized Arbacia eggs. The values of \( k \), the velocity constant, of three separate experiments represented by different symbols are plotted against the concentration, in per cent, of sea water. A sigmoid curve is thus obtained. (Each point represents the mean of six cells.)](attachment:image.png)

\[ \frac{(a - x)}{a} = \log \frac{x}{a} \]

per cent, 0.028; from 70 per cent, 0.028; from 60 per cent, 0.026. The differences
are not regarded as significant. The results are plotted in Fig. 1. In each case
the observed points from the several experiments fall along a single curve calcu-
lated from the equation \( kt = \log \frac{a}{a - x} \), as described in the legend. It is seen
that the curves for cells taken from 60, 70 and 80 per cent sea water are merely
upper segments of the curve for cells taken from 100 per cent sea water.
Similar results were obtained on causing cells to shrink on being transferred from various solutions into 100 per cent sea water at 19°C., i.e. the values of the velocity constants were practically the same, though more variation was encountered. The results were as follows: from 40 per cent, \( k = 0.15 \) from 50 per cent, 0.13; from 60 per cent, 0.14; from 70 per cent, 0.12; from 80 per cent, 0.15.

This point established, the problem was greatly simplified. We were justified in causing cells to swell or shrink in any convenient concentration of sea water, and then transferring them to any other concentration, the effect of which we wished to study.

We were therefore able to carry out the following experiments on the effect of concentration: cells were transferred from 100 per cent sea water into 20, 40 and 60 per cent sea water respectively. The effect of 80 per cent sea water was studied by first causing cells to shrink in 125 per cent, then to swell in 80 per cent. The effects of 100 and 125 per cent sea water were determined by first allowing cells to swell in 50 per cent and then transferring them to 100 and 125 per cent.

A large number of experiments were done of which three, represented in Fig. 2, covered the entire range of concentrations. When the velocity constants are plotted against concentration of sea water in per cent, a sigmoid curve is obtained. It is seen that the value of the velocity constant in 125 per cent sea water is more than ten times as great as in 20 per cent, at the same temperature.

These experiments were carried out at 15°C. Other experiments at different temperatures (from 12° to 24°C.), covering parts of the concentration range, also give an S curve at all the temperatures employed.

DISCUSSION.

The experiments show that the velocity constant of the diffusion process is altered by change in the concentration of the medium. As the medium is made more dilute (by the addition of distilled water), the velocity constant becomes smaller, and, conversely, increase in the concentration of the medium leads to increase in the velocity constant (Fig. 2). At any concentration, the rate of the process still obeys the fundamental diffusion equation, \( kt = \ln \frac{a}{a-x} \). But the

\(^3\) The eggs had not reached equilibrium in 40 per cent sea water.
The fact that the value of \( k \) depends on the concentration of the medium, points to another factor modifying the rate of diffusion. This factor would seem to be a change in permeability of the membrane to water. It appears that dilution of the medium decreases permeability to water, while concentration increases it.

The mechanism involved may be pictured as follows:

Let us assume that water diffuses into and out of the cell through pores in the membrane, and that the size of these pores varies with the concentration of the medium, in the sense that they become larger the greater the concentration of the medium. Such a condition might be explained by regarding the cell membrane as a partially hydrated gel, which takes up water from hypotonic solutions, and gives off water in hypertonic solutions. In the one case, the gel can be imagined to swell and so reduce the size of its pores, in the other case to shrink so that the pores become more widely opened, and thus allow more rapid diffusion of water. The membrane can be thought of as permeated by the outside solution and instantly coming into osmotic equilibrium with it.

The \( S \) shape of the concentration effect can now be explained by the further assumption that the hydrophilic membrane takes up water at first to a great extent as the concentration of the outside sea water is decreased, but then to a less and less extent as the sea water is further diluted and the membrane approaches the saturation point. Conversely, the same reasoning can be applied when the concentration of sea water is increased—at first the membrane yields its water readily but at greater concentration only to a small extent.

This hypothesis therefore states that the lower the osmotic pressure of the sea water, the less permeable is the membrane to substances passing through its pores, and conversely. Such a theory is apparently capable of being tested experimentally.

**SUMMARY.**

1. Using the unfertilized egg of the sea urchin, *Arbacia*, as osmometer, it was found that the rate with which water enters or leaves the cell depends on the osmotic pressure of the medium: the velocity constant of the diffusion process is higher when the cell is in concentrated sea water, and lower when the sea water medium is diluted with dis-
tilled water. Differences of more than tenfold in the value of the velocity constant were obtained in this way. When velocity constants are plotted against concentration of medium, a sigmoid curve is obtained.

2. These results are believed to indicate that cells are more permeable to water when the osmotic pressure of the medium is high than when it is low. This relation would be accounted for if water should diffuse through pores in a partially hydrated gel, constituting the cell membrane. In a medium of high osmotic pressure, the gel is conceived to give up water, to shrink, and therefore to allow widening of its pores with more ready diffusion of water through them. Conversely, in solutions of lower osmotic pressure, the gel would take up water and its pores become narrow.

BIBLIOGRAPHY.