Biochemistry. — *Tissues of prismatic celloidin cells containing Biocolloids.* VII. *Stagnation effects.* By H. G. BUNGENBERG DE JONG and B. KOK. (Communicated by Prof. H. R. KRUYT.)

(Communicated at the meeting of January 31, 1942.)

In communication V of this series the effects were studied on the complexcoacervate gelatine + gum arabic of a number of salts and non-electrolytes added to the 0.01 N acetic acid[1]). The effects occurring when the new medium is led continuously past the membrane (inflow effects) have been described in that communication, likewise the effects occurring when after that 0.01 N acetic acid is continuously led past the membrane (outflow effects). In some substances added to the 0.01 N acetic acid it was seen that some special effects are obtained, when the tap of the reservoir containing the inflowing liquid is closed. As these effects are only the consequence of the stagnation of the liquid flowing past the membrane, they were called *stagnation effects*. They have been observed in 5/9 mol glucose, saccharose, glycerine and 20 m. aeq. KCl; but the interpretation which we shall give below makes us expect these effects to be far more general. That we cannot further observe them is possibly owing to the fact that the in- and outflow effects often take place with such a rapidity and intensity, as to render the observation of the comparatively weak stagnation effects very difficult.

We will here give a short description of the stagnation effects with 20 m. aeq. KCl. After the coacervation with 0.01 N acetic acid has been brought about and the parietal coacervate contains only few little vacuoles, we change to 0.01 N acetic acid + 20 m. aeq. KCl. We then see the little vacuoles left in the parietal coacervate disappear and when we continue to lead this medium past the membrane the result is the inflow vacuolization described in communication V. We do not, however, wait for this result, but turn off the tap of the reservoir, then we see a *new vacuolization arise* (many little vacuoles) [2]), which decreases when the tap is quickly turned on again [3]). This *stagnation effect* consisting of vacuolization which decreases when the medium liquid is made to flow again, can be repeated a few times, but after some time the effect becomes less intensive finally not to occur at all. So the *stagnation effect* is a phenomenon occurring only at the beginning of the inflow period and is apparently connected with the fact that medium and contents of the cells are not yet in equilibrium. Figure 1 illustrates our interpretation of the *stagnation effect*: in this figure the object glass against which the celloidin membrane is located is shaded and the border between membrane and the adjacent medium flowing past it is indicated by a vertical line. Apart from the lumen of the cell the space in between contains 1. celloidin wall on the right, 2. celloidin wall on the left and 3. stagnating liquid space between membrane and object glass. But as these are also accessible to the diffusing substance these details have no fundamental significance for us and therefore we have indicated all this as cell in the figure.

We now set out on the ordinate the concentration of the substance added to the medium. In the medium liquid flowing rapidly past the membrane this concentration may be taken as constant and is therefore represented by a horizontal line from a, the celloidin wall, to the right. A short time after the onset of the inflow there is in the cell a certain quantity of

---

1) H. G. BUNGENBERG DE JONG and B. KOK, Proc. Ned. Akad. v. Wetensch., Amsterdam, **45**, 67 (1942).

2) Any vacuoles that may still be present then disappear rapidly.

3) Simultaneously with the decrease of the vacuoles formed on stagnation a new generation of vacuoles belonging to the normal inflow effect may be formed.

the diffusing substance, the concentration of which decreases rapidly to the left of the celloidin wall. The course of this concentration is indicated in fig. 1 by straight lines for the sake of simplification.

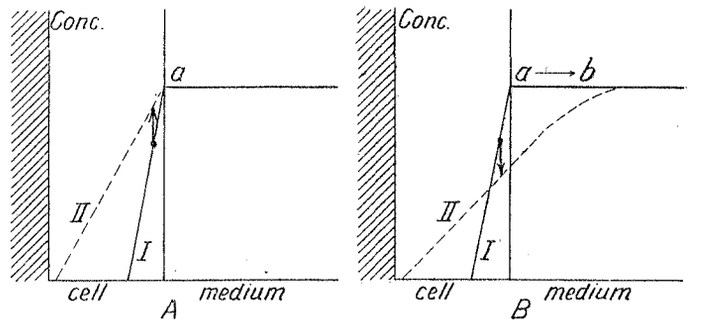

Fig. 1.

Incidentally we note that this consideration gives us at once an explanation of the detail mentioned in Communication V, that a new vacuolization always begins deep in the coacervate, i.e. on that side of the coacervate which is turned to the medium liquid flowing past the membrane. For this zone of the cell always comes first into contact with the diffusing substance coming from the medium. Moreover the concentration change takes place much more rapidly here than more to the left in the cell, which always promotes the occurrence of vacuolization.

For the explanation of the stagnation effect we now compare A and B in fig. 1. We suppose that line II in A shows the situation some time later. It is clear that at any depth in the cell the concentration of the substance diffusing inwards only increases in the course of time from the onset of the inflow until equilibrium has been reached.

In figure 1 B we shall now see what happens when we stop the flow of the medium liquid. Now there is no longer a liquid flowing past the membrane which preserves the concentration of the diffusing substance at a constant value, practically to the celloidin wall (a). As the diffusion continues, a certain zone of the stagnant medium (a—b) becomes poorer in diffusing substance, so that after some time the concentration course will be represented by line II in B. It is seen that in a zone of the cell lying close against the celloidin membrane, the concentration of the substance diffusing inwards has not increased, but decreased (compare the two arrows in fig. 1 A and B). Thus we arrive at the conclusion that *stagnation effects* are practically *local outflow effects,* with which the fact is in accordance that with glucose resp. 20 m. aeq. KCl vigorous and rapid vacuolization takes place on outflow. It also stands to reason that stagnation effects can be well observed exactly after a short period of inflow, that they are less distinct after longer inflow and finally do not occur at all after a sufficiently long period of inflow. Finally it is also clear that as there is here only a temporary local and not very great decrease of the concentration, the vacuolization is only weak in the stagnation effect and therefore possibly escapes observation in less favourable substances than KCl and glucose. Stagnation effects on outflow have not yet been observed by us. They ought also to occur, but possibly the circumstances are even less favourable here than on inflow.

Leiden, Laboratory for Medical Chemistry.