Micro-Electrode Penetration of Ascites Tumour Cells

EHRLICH ascites tumour cells from mice were suspended in Krebs phosphate Ringer in agar and maintained at 37° C. in a small chamber. Ling–Gerard micro-pipettes were filled with 3 *M* potassium chloride. They had a tip resistance of 30–50 megohms, and were checked for tip potentials by Adrian's method[1]. Any showing tip potentials greater than 2 mV. were discarded. Penetrations were observed microscopically under a magnification of 600.

The input valve ('Hivac' *XE*2) and micro-pipette holder were carefully insulated from earth, the measured input resistance being greater than 10^{11} ohms. The grid current was less than 10^{-13} amp., the valve being biased above contact potential so that with the grid open, or the tip of the micro-pipette deliberately blocked by pushing it into the 'Vaseline' lining the bath, a negative potential resulted.

In general, a difference of potential of 5–10 mV., positive inside, appeared when the microtip just penetrated the cell wall. This difference of potential was stable and could be obtained repeatedly on successive impalements of the same cell. The microtip could be moved about inside the cell for a small distance without changing this difference of potential.

On pushing the microtip farther into the middle of the cell the difference of potential changed abruptly to a value of 20–40 mV., negative inside. This difference of potential remained constant for about 10 sec. and then declined slowly; small movements of the tip caused no further change. If the tip was withdrawn after one or two seconds, the sequence just described could be obtained several times in one cell.

Under favourable conditions the following sequence of events is observed. On bringing the microtip up to the cell a slight dimpling of the outer wall is seen. As penetration occurs this dimpling disappears and the cell seems to seal itself around the tip, as usually occurs with other types of tissue. At this stage a difference of potential of 5–10 mV. appears abruptly. As the tip is moved farther into the cell, an invagination appears in the interior granular material. This suddenly disappears and at the same time the potential difference changes abruptly to 20–40 mV., negative inside. In some cells the negative difference of potential appeared immediately on penetration. In others only the positive difference of potential was obtained, but in most of these the negative difference of potential appeared on a second penetration in a slightly different direction.

It would seem that the initial positive difference of potential exists between the cytoplasm and the external medium and that the negative difference of potential occurs on penetration of some internal structure of the cell.

Electron micrographs[2] show that the ascites tumour cell, the diameter of which is about 20–30μ, contains a very large nucleus, which may occupy up to half the volume of the cell and is often situated eccentrically; and that as the cells deteriorate they may develop vacuoles almost as large. However, with cells that were obviously deteriorating, the negative difference of potential was elicited much less frequently (although the positive could usually be obtained). Thus it is probable that the structure penetrated was the cell nucleus.

The positive internal potential of the cytoplasmic membrane could be explained, on the basis of the Goldman equation, if one assumes a high permeability to sodium compared with potassium[3]. It has been shown[4] that these cells have an abnormally high permeability to sodium.

I wish to thank Prof. F. H. Shaw for his encouragement and helpful advice during the course of this work.

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Differences between Self-Incompatibility and Self-Sterility

THE term 'self-sterile' is used for describing plants which fail to set self-seed when they are grown in isolation or when they are self-pollinated. Silow[1] reviewed the early usage of this term and proposed the term 'voluntarily self-sterile' to describe *Lotus* plants that fail to set seed when isolated from insects and 'artificially self-sterile' when they still fail to set self-seed following artificial self-pollination. He observed that several *Lotus* species in isolation set more self-seed following artificial self-pollination than the same plants without artificial self-pollination. This difference can be accounted for by the presence of a stigmatic membrane which prevents germination and growth of any pollen. This membrane is ruptured by artificial self-pollination or by insect pollination of the flower, after which pollen can germinate and grow. The presence of a stigmatic membrane was observed by Elliott[2] in *L. tenuis* and by Giles[3] in *L. corniculatus*. Giles[3] was unable to observe any difference in germination between self- and cross-pollen once the membrane ruptured. The presence of a similar membrane was observed by me[4] in several other *Lotus* species. This membrane can account for the observed voluntary self-sterility and for differences between voluntary and artificial self-sterility, but it does not account for artificial self-sterility.

Several mechanisms which can cause artificial self-sterility are known. One of these is genetic 'self-incompatibility', as was first described by East and Manglesdorf[5]. Another is 'somatoplastic sterility' which causes post-fertilization abortion, first described by Brink and Cooper[6]. Several other possible causes have been postulated. Mechanisms which involve post-fertilization abortion lead to destruction of individual ovules, whereas incompatibility saves the ovules for fertilization by any cross-pollen that may be present. Post-fertilization abortion may be suspected if all ovules do not form seeds following pollination by a mixture of cross- and self-pollen. However, variability in rate of ovule development within ovaries may be expressed in the same way, as discussed by me[4].

The terms 'self-incompatibility' and 'incompatibility' are used for describing certain genetic outbreeding mechanisms of a type first described by East and Manglesdorf[5]. Two main systems are found in angiosperms with homomorphic flowers. These systems are controlled by incompatibility alleles at one or two loci in which incompatibility is found