The Behaviour of the Golgi bodies during nuclear division, with special reference to Amitosis in Dytiscus marginalis.[1]

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With 4 Text-figures.

CONTENTS.

1. INTRODUCTION.

DICTYOKINESIS, or division of the Golgi bodies during cell division, has recently been worked out by Professor J. Brontë Gatenby and the present writer in the male germ-cells of several animals (**5**). It was found that the distribution of the Golgi elements, or dictyosomes, to both halves of a dividing cell was a very haphazard process and was unaccompanied by any splitting of the dictyosomes such as occurs in the case of the chromosomes. In the cricket Stenobothrus, the dictyosomes

[1] Part of the materials used in this research was purchased by a Government Grant of the Royal Society, for which I express my thanks,

become scattered in the cytoplasm of the spermatogonium before cell division takes place, and they remain in this condition during meiosis, so that approximately a half become contained within each of the newly-formed cells. A different process, however, occurs in the Mammals, Mus and Cavia, and in the Molluscs, Helix and Limnaea. The Golgi bodies in the spermatogonia of these types consist of a number of dictyosomes arranged around the archoplasm, inside which is the centrosome. As this latter organ divides, preparatory to the formation of the spindle, its two constituent parts separate and carry with them to both ends of the cell, approximately half of the archoplasm, still with the dictyosomes attached. During late prophase, the dictyosomes become temporarily detached from the archoplasm and scattered throughout the cell, and then at the late telophase they collect together again around the archoplasm.

The examples of dictyokinesis described in our previous paper were those which occurred concurrently with meiotic nuclear division. Professor Gatenby suggested to me the desirability of investigating the behaviour of the Golgi body during amitotic nuclear division, and in the present paper is described the behaviour of the apparatus during amitosis in the follicle cells of the ovary of the beetle, Dytiscus marginalis.

2. Previous Work.

Deinecka (**1**) has described dictyokinesis in the dividing epithelial cells of Descemet's membrane and connective-tissue cells of the cornea, during both mitotic and amitotic nuclear division. He found that the Golgi body surrounded the archoplasm, and during mitosis divided into two parts so that each daughter-cell received a 'Netzapparat', as he calls it, but that in amitosis there is no division of the centrosome and no change in the Golgi body. These observations are quoted by Macklin (**8**) in support of his own conclusions derived from a study of nuclear division in cells of tissue cultures of the heart of the embryo chick, that amitosis involved

merely division of the nucleus, and not of the cytoplasm. He observed that binucleate and polynucleate cells were formed as the result of amitotic nuclear division. During such division the centrosome and archoplasm remained unchanged. The archoplasm could be seen in the living cell, but not the centrosome; but the latter was to be seen in fixed preparations stained with iron haematoxylin as two small black bodies embedded in the archoplasm. Mitochondria were visible in the living cells, but not the Golgi body.

Macklin's observations are of special interest in that they substantiate materially the evidence that has been accumulated against the view upheld by writers such as Meves (**9**), that mitosis can occur in an amitotically-formed nucleus. Binucleate cells were observed by Macklin to undergo mitosis, but in this process the nuclei which had been formed by amitotic division came together and their chromatin masses fused to form the chromosomes which underwent the usual stages of mitotic division. It is therefore concluded that amitosis does not imply division of the cytoplasm but only fission of the nucleus.

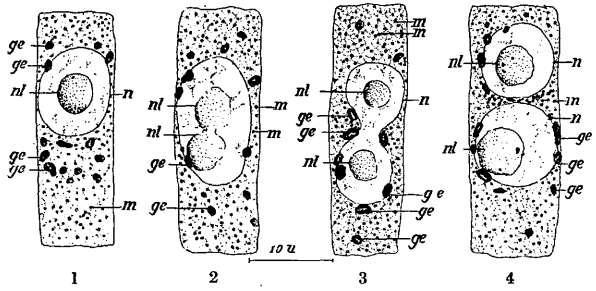
3. The Golgi Body during Amitosis in the Follicle Cells of the Ovary of Dytiscus.

In common with most insects Dytiscus has an ovary laterally disposed on either side of the abdomen. Each ovary is composed of a number of tubules containing a single row of oöcytes in all stages of development, the most mature being at the distal end. The oöcytes are surrounded by the follicle cells, and between each oöcyte is a group of nurse or nutritive cells whose function is to provide nourishment for the developing oöcytes. At the proximal end of the ovuliferous tubules is a mass of undifferentiated cells from which arise three types of cells, viz. oöcytes, nutritive cells representing modified oöcytes, and the follicle cells in which the behaviour of the Golgi body during amitosis was studied.

It was found rather difficult to impregnate the body in the follicle cells of insects. After a number of unsuccessful attempts

the best results were obtained by adopting Cajal's method (**4**) with slightly longer fixation than he describes. Ovaries of Dytiscus were fixed for about eight hours in Cajal's standard fixative, and after rapid washing in distilled water were left in silver nitrate solution for three days. Prepared slides were stained by safranin or carmine. The Golgi body then appears as a number of black granules in a pink-coloured cytoplasm, the mitochondria when visible are usually brown in colour, and the nucleoli of the cells are red.

TEXT-FIGS. 1–4.



Follicle cells of the ovary of Dytiscus marginalis. The upper cell-wall is in each case in contact with the oöcyte wall. *ge*, elements of the Golgi body (Dictyosomes); *m*, mitochondria; *n*, nuclear membrane; *nl*, nucleolus (plasmosome).

In the text-figures are shown the various stages of amitosis. At fig. 1 is seen the so-called 'resting stage' of the cell. It will be observed that there is a single nucleolus within the nucleus, and scattered through the cytoplasm are the darkly-impregnated elements of the Golgi body, while the mitochondria are more or less evenly distributed in the cell. In the stage shown in fig. 2 the nucleus has elongated and the nucleolus is dividing into two. The Golgi elements still remain scattered in the cytoplasm, but it will be noticed they show a tendency to lie near the nuclear membrane—a tendency which is apparent in the other figures. At a later stage, as shown in fig. 3, the

two parts of the nucleolus have separated and the nucleus is greatly constricted, but the dictyosomes are still irregularly scattered: while in the cell shown in fig. 4, when the two parts of the nucleus are completely separated, the dictyosomes are still irregularly disposed around them.

It will be noticed that the nucleolus appears to play quite an important part in this process: its division seeming to initiate the division of the nucleus. This process has been verified by observations on material prepared by fixation in corrosive acetic and Bouin, and stained with Mann's methyl blue eosin (**7**). In such preparations the nucleolus stains oxyphil, and is apparently of the nature of a plasmosome. Its appearance is the same in both kinds of preparations. This type of amitosis, originally described by Remak, in which the nucleolus appears to play an important part, has been found by recent workers to be exceptional rather than typical, and Macklin, observing amitosis in living cells, says 'the division of the nucleolus has no direct relationship to nuclear division. It may, however, have to do with the size of the nuclear portions' (**8**).

The extent to which the dictyosomes are distributed in the resting follicle cell is subject to variation. In some cases, evidently owing to the large size of the nucleus in comparison with the width of the cell, the elements of the apparatus are crowded together towards its outer wall and appear in rare cases to be attached to an archoplasm; but this does not occur when the cell is dividing amitotically, and in no case has the separation of two distinct groups of dictyosomes, as occurs in mitosis, been observed.

4. Discussion.

Gatenby has suggested (**3**) that the scattering of the Golgi elements during oögenesis is a means whereby it is able to exert a maximum formative influence upon the cytoplasm, as well as prepare for even distribution in the cells of the segmenting ovum. In a previous paper (**6**) I have described how the dispersing dictyosomes influence the formation of

yolk in the oöcyte of the Mollusc Patella. It would seem possible, therefore, that the spreading out of the apparatus in the follicle cells of an insect might be related to the high degree of metabolism existing in such cells.

Chun, quoted by Nakahara, regarded the division of the nucleus in amitosis as a means of increasing the nuclear surface as an aid to metabolic interchange between nucleus and cytoplasm; while Flemming pointed out that amitosis was especially associated with intense secretive and assimilatory activity, but he considers such cells as being on the way to degeneration (**2**). Recent work has shown that fragmentation of the nucleus does occur in pathological growths, in cells subject to faulty nutritive conditions, and in tissue cultures which have been left unattended for some time. Such fragmentation is regarded by Macklin (**8**) as an altogether different phenomenon from amitosis, but in the past there is no doubt that there has been confusion between the two.

Nakahara, who has made an investigation into the subject of amitosis in adipose cells of insects and an extensive survey of the literature of the subject, concluded that 'amitosis, occurring in secreting or reserve forming cells and in other cells of similar activity, may be for the purpose of securing an increase of the nuclear surface to meet the physiological necessity due to the active metabolic interchanges between the nucleus and cytoplasm. Apparently it is not a method of cell multiplication nor a sign of degeneration or senescence of cells, but, whenever it occurs, it seems to indicate an intense activity in the vegetative functions of the cell' (**11**). It is altogether in accordance with our present knowledge of the Golgi apparatus to assume that in such cells, as for example the follicle cells of insects' ovaries, the dictyosomes scattered in the cytoplasm would play a by no means unimportant part in the lipoid metabolism.

In conclusion, I have to acknowledge my indebtedness to Professor J. Brontë Gatenby, of Trinity College, Dublin, for his kindness in reading through the manuscript of this paper.

5. Conclusion.

We may now recognize the following modes of behaviour of the Golgi bodies during nuclear division:

(1) During karyokinesis the Golgi bodies may either,

(*a*) remain scattered in the cytoplasm and be approximately shared out amongst the two newly-forming cells, e.g. male germ-cells of Stenobothrus (**5**);

(*b*) divide into two masses surrounding the separating centrosomes and thus pass into each cell, e.g. (i) during meiosis in the male germ-cells of the Molluscs, Helix and Limnaea, and the Mammals, Mus and Cavia (**5**); (ii) during mitosis in the epithelial cells of Descemet's membrane and connective-tissue cells of the cornea (**1**).

(2) During amitosis either they

(*a*) remain as a number of elements or dictyosomes arranged around the archoplasm, e.g. in mammalian epithelium (**1**), or they

(*b*) become irregularly scattered throughout the cytoplasm, as described in this paper in the follicle cells of insects' ovaries.

It is suggested that these differences are related to different conditions of metabolism existing in cells exhibiting these phenomena.

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