Incidentally, these more complicated cases serve to underline the importance of avoiding ambiguity between the designations for the phenotypes and genotypes.

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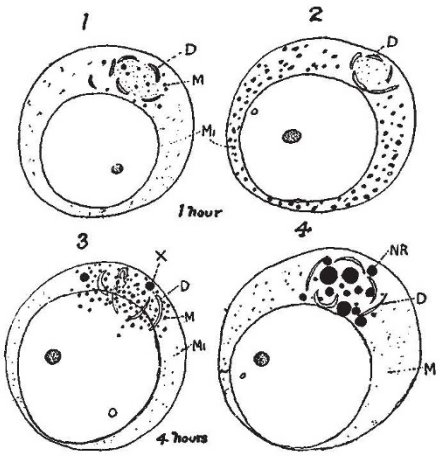
Methylene Blue as a Vital Stain for the Golgi Apparatus

EXPERIMENTS have been made with five specimens of methylene blue as a vital stain for the spermatocytes of *Helix aspersa*. The specimens were an old German 'polychrome methylene blue', and four others, 'pure methylene blue', and just 'methylene blue'.



In a suitable concentration all the specimens acted in the same way, being segregated into small droplets (*M*) inside and outside the Golgi apparatus (*D*), neither the mitochondria, M_1, nor the lenticular dicytosomes staining (Fig. 1). At a higher concentration of the dye (not the polychrome methylene blue) the mitochondria stained intensely, rivalling the best Janus green vitally stained cells (Fig. 2). In many of these cases the methylene blue granules associated with the Golgi apparatus (Fig. 1) did not appear. The polychrome methylene blue at these higher, perhaps lethal, concentrations tended to stain the mitochondria blue and rarely the dictyosomes of the Golgi apparatus a reddish colour. The main difference between methylene blue and neutral red as vital dyes for *Helix* spermatocytes is that the methylene blue can be made to stain mitochondria or to segregate into droplets in the region of the Golgi apparatus, whereas neutral red does not stain the mitochondria or Golgi dictyosomes, but easily segregates into droplets which grow in size according to the length of time the cells are exposed to the dye. In Fig. 3 is a cell which has been exposed to a weak methylene blue solution for four hours. The number of methylene blue segregation artefacts has increased very greatly, the dictyosomes are still unstained and in rare cases the methylene blue segregation cavities may be large (*X*). Generally speaking, these cavities do not become as large as the neutral red artefacts (Fig. 4, after four hours).

It is interesting to note that the methylene blue segregation artefacts shown in Fig. 3 are spread both inside and well outside the region of the Golgi apparatus. In more highly differentiated cells, such as nerve or salivary cells, the behaviour of the methylene blue dye will be more complicated.

It is hoped to publish the results of these and other experiments on methylene blue and Sudan Black B elsewhere in a fuller form.

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Passage of Hæmoglobin from Blood into Eggs of *Daphnia*

IT is known that the amount of hæmoglobin dissolved in the blood of *Daphnia* varies inversely with the oxygen content of the water in which the animals live. The changes in concentration of the hæmoglobin have been studied by a quantitative method which allows measurements to be made on a single *Daphnia*: the colour of the animal's blood is compared under the microscope with known dilutions of human blood[1]. In the course of further work involving the use of this method it was noticed that parthenogenetic females with eggs in the brood pouch have less hæmoglobin in their blood than those with embryos in the pouch. The difference is greatest in populations producing numerous young.

The parthenogenetic eggs of *Daphnia* contain hæmoglobin[2]. It can be seen under the microscope with reflected light that the ovaries become progressively pinker as they enlarge during the last few hours preceding egg-laying. The microspectroscope shows that this pink colour is that of oxyhæmoglobin. Moreover, at the same time as the pink colour is increasing in the ovaries, the blood becomes paler. This decrease in the hæmoglobin content of the blood may amount to one-third. The inference is that hæmoglobin passes from the blood into the eggs as they develop in the ovary. The course of the decrease during the six hours before the moult, which immediately precedes egg-laying, has been studied quantitatively in a number of individual *Daphnia*. It is at first gradual and then rapid. Part of the final decrease in the pink colour of the blood is due not to a loss of hæmoglobin but to a dilution of the blood resulting from the moult and the increase in body-size which is its consequence. This dilution, however, accounts only for a small proportion of the decrease in blood colour. After egg-laying, the hæmoglobin of the blood is gradually regenerated to its original level. The course of this regeneration has been followed quantitatively over the 2–3 days during which the embryos develop in the brood pouch and are finally liberated prior to the next moult.

The molecules of *Daphnia* hæmoglobin appear to be of two sizes, with weights respectively one-half and six times that of mammalian hæmoglobin; the