

DNA and heterochromatin cycles in male gamete development of *Oedogonium*

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Abstract

In antheridial nuclei of *Oedogonium cardiacum* (Hass.) Wittr. during male gamete formation, the DNA presynthetic period is shortened, the synthetic and postsynthetic periods are lengthened (G_1 — 15%, S — 58%, G_2 — 29%) as compared with those in vegetative cells (G_1 — 31%, S — 44%, G_2 — 25%). Differences in the ratio of the relative amounts of eu- and heterochromatin in the antheridial mother cell (AMC) nucleus and antheridial cell nucleus were determined. Hydrolysis curves revealed two peaks of higher optical density (3.5 and 8.5 h hydrolysis with 5N HCl at 23°C), corresponding to eu- and heterochromatin. Heterochromatin in antheridial cell nuclei was characterized by a greater optical density than that of heterochromatin in AMC nuclei. The presence of two local optimal density maxima is considered as facultative and constitutive increasing of chromatin condensation in the process of male cell development.

INTRODUCTION

The functional aspects of the male gametes development are studied rather insufficiently, contrary to the morphology and developing male gametes investigations. The main task of the paper was the mitotic cycle of the developing male gametes and changes in the relative amounts of the diffuse and condensed chromatin at the final stages of them.

MATERIALS AND METHODS

On filamentous green alga — *Oedogonium cardiacum* (Hass.) Wittr. — the average amount of DNA in the spermatozoid nucleus was determined by application of variation statistics to the mass of nuclei from these cells. 50 pairs of chromosomes served as the standard of ploidy. The histogram asymmetry coefficient was determined by the third order momentum. When plotting curves of DNA synthesis in nuclei from antheridia and vegetative cells, measurements of 500 nuclei of each type of cells were used.

To determine the relative amounts of diffuse and condensed chromatin, the method of 5N HCl fractional acid hydrolysis of different duration (from 10 min till 14 h at 23°C) was used in all 35 terms with subsequent staining of nuclei by the Schiff reagent. After Feulgen reaction the DNA content in the nuclei was measured by the two-wave method in one-beam cytospectrophotometer with 500 and 578 nm wave lengths.

RESULTS AND DISCUSSION

The measurement of 100 spermatozooids permitted to determining their DNA content (6.74 ± 0.18 relative units). The low coefficient of asymmetry (3.5%) and the fact that the histogram (Fig. 1) is subjected to normal distribution is evidence for the absence of replication.

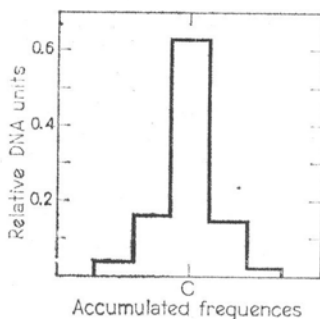


Fig. 1. Amount of DNA in spermatozoid nuclei of *Oedogonium cardiacum*.

In the process of the male gamete formation in *Oedogonium cardiacum* three types of cells are produced: the material cells of antheridia (MCA), antheridia and spermatozooids. Simultaneously, a decrease in cell size, decrease in the amount of cytoplasm in the cells and intensification of nuclear heterochromatinization are observed.

Data on DNA synthesis in vegetative cells (G_3 — 31%, S — 44%, G_2 — 25%) and antheridia (G_1 — 15%, S — 58%, G_2 — 29%) (Fig. 2) are evidence for shortening of the duration of the presynthetic period and an increase in the duration of the synthetic and postsynthetic periods. The prolongation of the S period may be connected with the fact that more chromatin is heterochromatinized in antheridial cells as compared with vegetative cells. Analogous changes in the cycle were traced in our laboratory in spermiogenesis in *Nicotiana tabacum* and *Lilium regale* (Bannikova et al., 1978) and are characteristic of spermiogenesis of a great number of animals (Dondua, 1967).

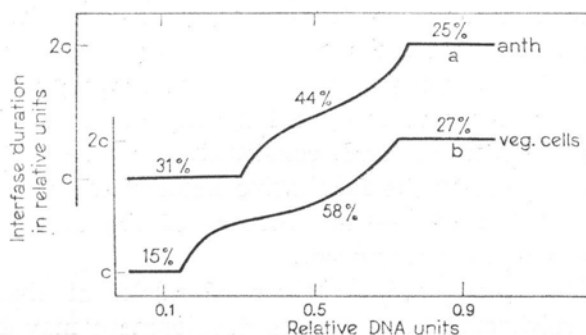
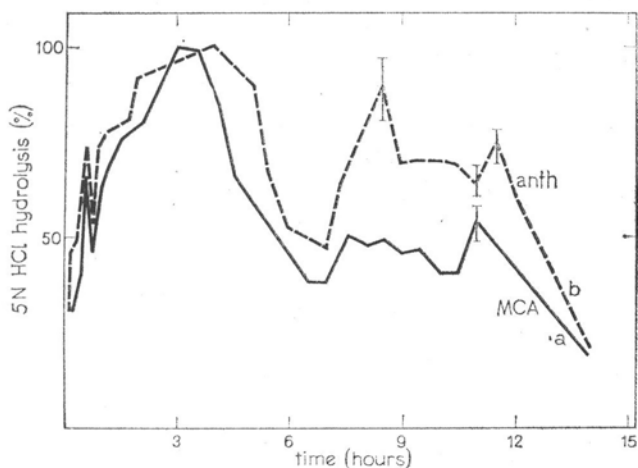


Fig. 2. DNA in antheridium nuclei (a) and vegetative cells (b) of *Oedogonium cardiacum*

Thus, differentiation of the male gametes is accompanied by morphological and biochemical changes. The peculiar proceeding of the cell cycle may be connected with changes in the nuclear cytoplasmic relation and decrease in cell size. The rate and sequence of synthetic processes proceeding the G_1 period antheridia cells, evidently, are the same as in MCA. Therefore, it is possible to suppose the existence of a correlation between a decrease in the antheridial cell size and the shortening of the duration of the presynthetic period in the cells.

While investigating the state of chromatin in the MCA and antheridial nuclei, the two-apex curves of hydrolysis were plotted (Fig. 3). The first apex in both curves corresponded to the diffuse part of chromatin, the second apex — to the condensed one. Basing on the curves



depending on the duration of 5N HCl hydrolysis at 23°C
Fig. 3. The relative content of DNA in nuclei of MCA (a) and antheridia (b)

of hydrolysis it is established trustworthily that an increase in the amount of heterochromatin areas occurs at the preceding the formation of spermatozooids. In segments of the curves corresponding to condensed chromatin two local maxima of optical density are marked, which may be considered as facultative and constitutive heterochromatin. In the antheridial nuclei a rise of the facultative heterochromatin share is seen clearly, and in MCA nuclei — an increase of the constitutive heterochromatin share is more pronounced.

Increase in the condensed structure of nuclei at the final stages of *Oedogonium cardiacum* male gamete development may be considered a result of facultative heterochromatization of chromatin areas previously being in the active diffuse state. A considerable amount of diffuse chromatin in antheridial nuclei may appear due to despiralization of the chromatin areas which earlier were in the condensed state and are now activated during the formation of the highly specialized male gamete possessing many new properties.

REFERENCES

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