Alternation of Generations and Nuclear Phases of *Ulothrix* flexuosa Kornmann prox. (Ulotrichales, Chlorophyceae)*1

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Alternation of generations and nuclear phases of *Ulothrix flexuosa* prox. cultured in incubators was examined. This alga was observed to exhibit dimorphic alternation of two generations, a haploid multicellular filamentous thallus and a diploid unicellular cyst. There were 8 chromosomes in vegetative cells of the filaments, in swarmers and gametes formed in filaments, and in swarmers in cysts. For cysts produced through conjugation of gametes, the number of chromosomes was 16. Meiosis of this species thus occurs in cysts at the time of swarmer formation.

1 Introduction

The genus, *Ulothrix*, is a green alga distributed widely in colder waters of Japan. The species *Ulothrix flexuosa* prox. (abbreviated as *U. flexuosa*) is commonly found from winter to spring in waters off southern Hokkaido. Kornmann⁴⁾ found the life cycle of *U. flexuosa* was to consist mainly of two generations. Although the life cycle of this species has been examined in detail, an adequate cytological explanation of the cycle has not yet to be made. The present study was thus conducted to clarify the nuclear phases of *U. flexuosa* with special attention to its meiosis.

2 Materials and Methods

Several filamentous thalli of *U. flexuosa* were collected off Muroran, Hokkaido in April, 1978. The filaments were rinsed in sterilized seawater to eliminate epiphytes and a few were then transferred to a PES medium⁹. They were maintained in incubators at 5°C under two different light-dark photoperiods; 9 and 15 hr (short-day) and 14 and 10 hr (long-day). Light intensity, provided by white fluoresent lamps, was ca. 5,000 lx.

For the cytological study, cultivated filaments were fixed in Zenker's fluid and stained by the Feulgen squash method². The number of chromosomes at each of the various

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cell stages was determined under a light microscope.

3 Results and Discussions

The present species was usually found adhering to rocks and stalks in the intertidal zone. Vegetative cells in the central parts of the filaments were 20-45 μ m in diameter and 10-15 μ m in length (Fig. 1, A). The chloro-

plasts were shaped like an incomplete ring with one pyrenoid.

Sporangia were formed in the vegetative cells of filaments under all conditions of culture. Each sporangium contained 4 to 32 swarmers (Fig. 1, B). The swarmers were generally pearshaped, having four flagella of equal length, one eye-spot, and a basal chloroplast with a single pyrenoid (Fig. 1, C). After settling, they germinated and developed into filaments.

Gametangia were formed in the same filaments as the sporangia were formed under

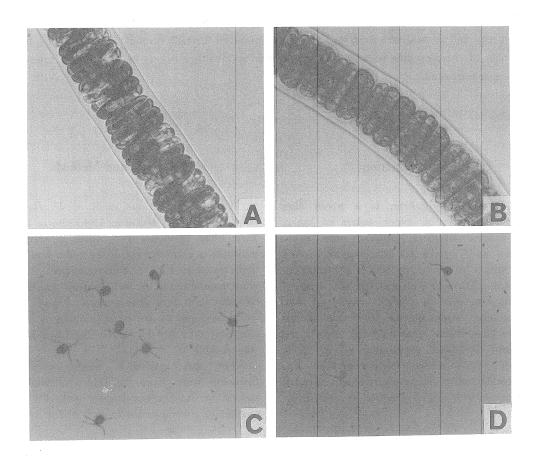


Fig. 1. Vegetative cells, sporanngia, swarmers and gametes in *Ulothrix flexuosa* Kornmann prox. A: vegetative cells, B: sporangia formed in filametous thallus, C: liberated swarmers stained by Gentian-violet. (A-D: x450).

the long day condition. Each gametangium contained 16 to 64 gametes (Fig. 1, B). The gametes were also pear-shaped but with two flagella of equal length and one eye-spot (Fig. 1, D). Sexual filaments were monoecious and sexual reproduction was isogamous (Fig. 2, A). Following sexual fusion, the zygotes settled onto the substrata (Fig. 2, B) and increased in size without cell division (Fig. 2, C-F); they

eventually formed thick-walled cysts in which 16 to 128 swarmers developed (Fig. 2, G, H). Following their liberation from the cysts, they formed the same shape as the swarmers in the filaments (Fig. 2, I). After settling, the swarmers began to germinate and grew into filaments. When there was no sexual conjugation, gametes developed parthenogenetically and showed zygote-type germination and

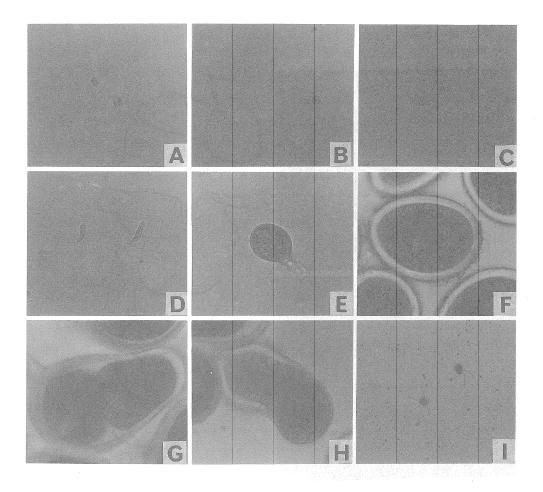


Fig. 2. Conjugation of gametes, zygotes and successive stages in the development of zygotes in *Ulothrix flexuosa* Kornmann prox. A: conjugation of gametes, B: settled zygotes, C-E: development of zygotes, F: thick-wall cysts, G, H: fertile cysts, H: formation of swarmers in cyst, I: liberated swarmers stained by Gentian-violet. (A-I: x420).

developed into thick-walled cysts.

In comparison with those described by Kornmann⁴⁾, the present specimens were characterized by having planospores in the cysts which were developed from zygotes.

Cytology

The chromosomes of the present species exhibited extreme contraction in the metaphase and anaphase (Figs. 3-5). There were 8 chromosomes in each vegetative cell of the

filaments (Fig. 3, A-D), in asexual swarmers (Fig. 4, A, B), and in gametes which produce sexual fusion (Fig. 4, C, D). Nuclear division occurred through mitosis, and meiosis was not be detected at any time during nuclear division.

Zygotes were attached to the substrata and developed cysts that increased in size without cell division. Each cyst contained one nucleus larger than that of vegetative cells. The characteristic stages of meiosis were observed at the initial division of the unicellular cysts.

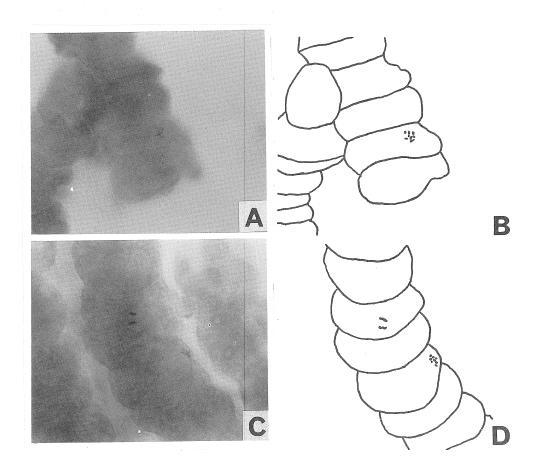


Fig. 3. Mitotic divisions in vegetative cells in *Ulothrix flexuosa* Kornmann prox. A: metaphase (n=8), B: drawing of the chromosome in A, C: metaphase and anaphase, D: Drawing in C. (A-D: x1,200).

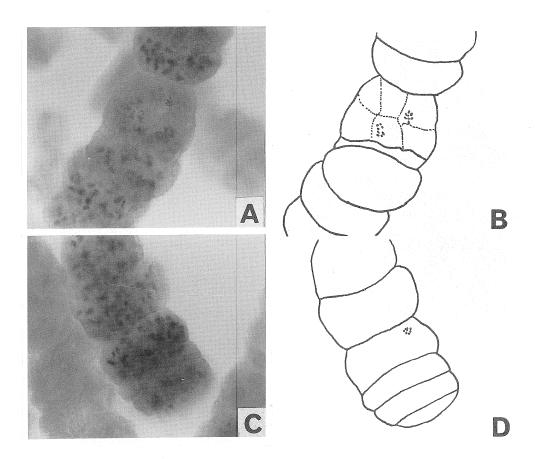


Fig. 4. Mitotic divisions in reproductive cells of filamentous thallus in *Ulothrix* flexuosa Kornmann prox. A: chromosome of swarmers (n=8), B: drawing of the chromosome in A, C: chromosome of gametes (n=8), D: drawing in C. (A-D: x1,200).

Each cyst possessed 16 chromosomes although some variations were noted in the number. Meiosis occurred during the first and second nuclear divisions of the cyst (Fig. 5, A-D), followed by several mitotic divisions. Each swarmer possessed 8 chromosomes and was haploid (Fig. 5, E, F).

Alternation of nuclear phases of the present specimens occurred between filaments and cysts. The filaments were haploid, while the cysts produced by the conjugation of gametes were diploid. However, swarmers that were formed in filaments and developed into filaments were haploid. This phenomenon in the case is referred to as a subcycle.

In other species of the genus *Ulothrix* studied, the number of chromosomes differed from that in the present species being 10 in *U. zonata*¹⁰, 5 in *U. rorida*⁶, and 14 in *U. subtillisima*¹⁰. Lind studied the vegetative cells and sporangia in the filaments of *U. rorida*. He concluded, in regards to the

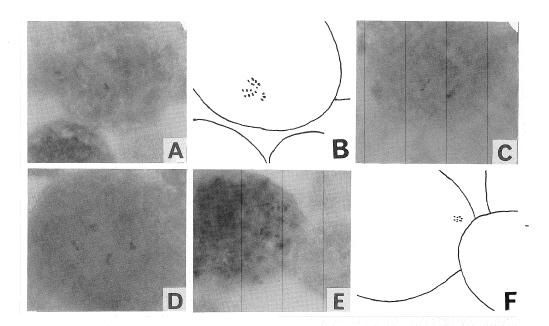


Fig. 5. Meiotic divisions in cysts at the time of swarmer formation in *Ulothrix fle-*xuosa Kornmann prox. A: metaphase I (2n=16), B: drawing of the chromosome in A, C: anaphase I, D: anaphase II, E: chromosome of swarmers in cyst (n=8), F: drawing in E. (A-E: x1,200).

alternation of nuclear phases that (1) filaments are haploid, (2) cysts formed by the conjugation of gametes are diploid, and (3) that meiosis occurs when swarmers are formed in cysts. The present data supports these conclusions.

Two asexual swarming phases were observed in the life cycle of *U. flexuosa*; each having exactly the same shape and number of chromosomes although their mode of nuclear division was quite different. New terms are therefore needed to distinguish these two kinds of swarmers. Swarmers formed in filaments through mitosis should be termed mitozoospores and those formed in cysts through meiosis, meiozoospores, as previously used in species of Blastocladiales in Fungi¹⁾. These terms should also be used for other species having the same life cycle such as *Urospora* spp. ^{35,8)} and *Capsosiphon fluvescens*⁷⁾.

Acknowledgments

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Ulothrix flexuosa Kornmann prox. の生活環と核相交代について

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室内培養実験から *Ulothrix flexuosa* Kornmann prox. の生活環を調べ、その核相交代を明らかにした。

本種は遊走子と配偶子を形成する単列細胞の葉体と配偶子の接合によって生じる Cyst 体との間に世代交代が行われていた。有性生殖は雌雄同株で同型配偶が行われていることが認められた。

核相については葉体が単相で Cyst 体が複相であることが認められ、減数分裂は Cyst 体に遊走細胞が形成される際に行われることが明らかになった。染色体数は葉体が n=8, Cyst 体が 2n=16であった。