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Alternation of Chromosomes in the Life History of *Laminaria japonica* Aresch.

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Abstract

Chromosomal alternations in the cycle of the life history of *Laminaria japonica* were observed. The chromosome number n=22 was counted in the first nuclear division in zoosporangium. It agrees with the previous count made by Abe of the present species in 1939. It was elucidated from the count of chromosomes that nuclei in gametophytes are haploid and those in sporophytes are diploid.

Introduction

The chromosomes of *Laminaria japonica* were counted and finely illustrated by Abe[1] in zoospore formation within the sporangium. Until now, many cytological studies of the Japanese species of Laminariales have been performed by the following investigators; Inoh and Nishibayashi[2], Kaneko[3], Nishibayashi and Inoh[4,5,6,7,8], Ohmori[9,10], Yabu[11,12,13,14], and Yabu and Tokida[15], but their works were all limitted to the area of zoospore formation, using the classical paraffine method in the same way as Abe's study[1] on *Laminaria japonica*. The recent papers reported by Evans[16] on seven species of British Laminariales, by Kemp and Cole[17] on *Nereocystis luetkeana*, by Cole[18] on *Macrocystis integrifolia*, by Robinson and Cole[19,20] on several species of *Alaria* from the West Coast of North America, each of them applied the aceto-carmine squash method, showed good results for the observation on chromosomes in gametophytes and young sporophytes and elucidated the chromosomal alternation between these generations. The present work was attempted to observe the chromosomal cycle in the whole generation of the life history of *Laminaria japonica* Aresch, by the use of the squash method.

Material and Method

Material of *Laminaria japonica* was collected in October 1970 at Usujiri, Oshima Province in Hokkaido. A part of maturing portion of the frond of the material was fixed in the evening of the day it was collected to observe the nuclear division in zoosporangium. The zoospores were liberated two days after collection and they were cultured on slides in vats containing Erd-Schreiber solution in a

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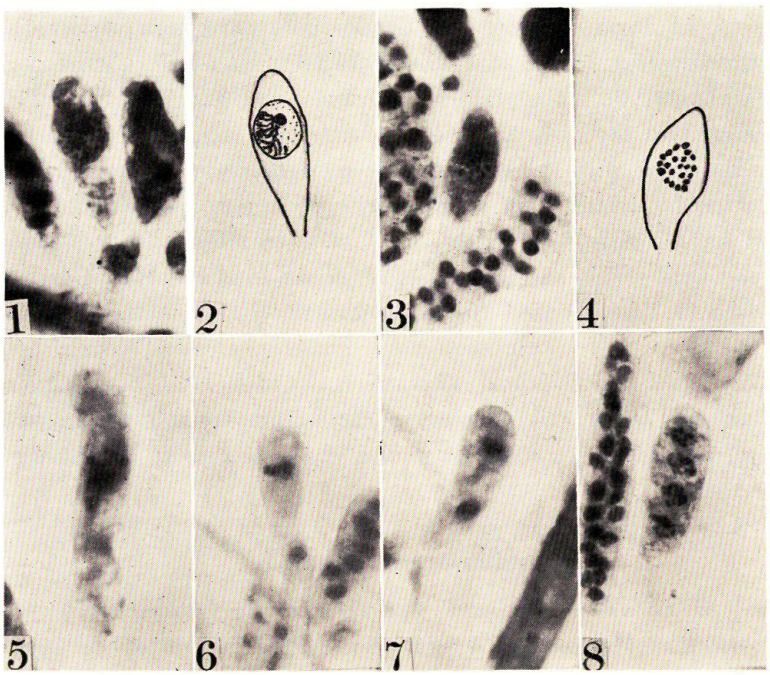
small culture room maintained at about 12°C and illuminated by fluorescent light in the Phycological Laboratory, Faculty of Fisheries, Hokkaido University at Hakodate. The slides, on which the germlings of zoospores had adhered, were taken from the culture vats and put into the fixative at intervals of five days. The maturing portions of the frond of the material and the germling of zoospores in culture vats were both fixed with acetic alcohol (1:3) solution and squash staining was done with Wittmann's aceto-iron-haematoxylin-chloral hydrate solution.[21)]

Results

Meiosis in the zoosporangium: The process of the nuclear division leading to the formation of zoospore within the sporangium is quite similar to that reported by Abe[1)]. The first nuclear division within a sporangium displays the prominent features of meiosis, in which the chromatin threads are gathering at one corner of the nuclear cavity at synapsis as shown in figs. 1 & 2, and bivalent chromosomes appear at diakinesis. In my study, chromosome count was only possible in late prophase and early metaphase of the first nuclear division, although a good number



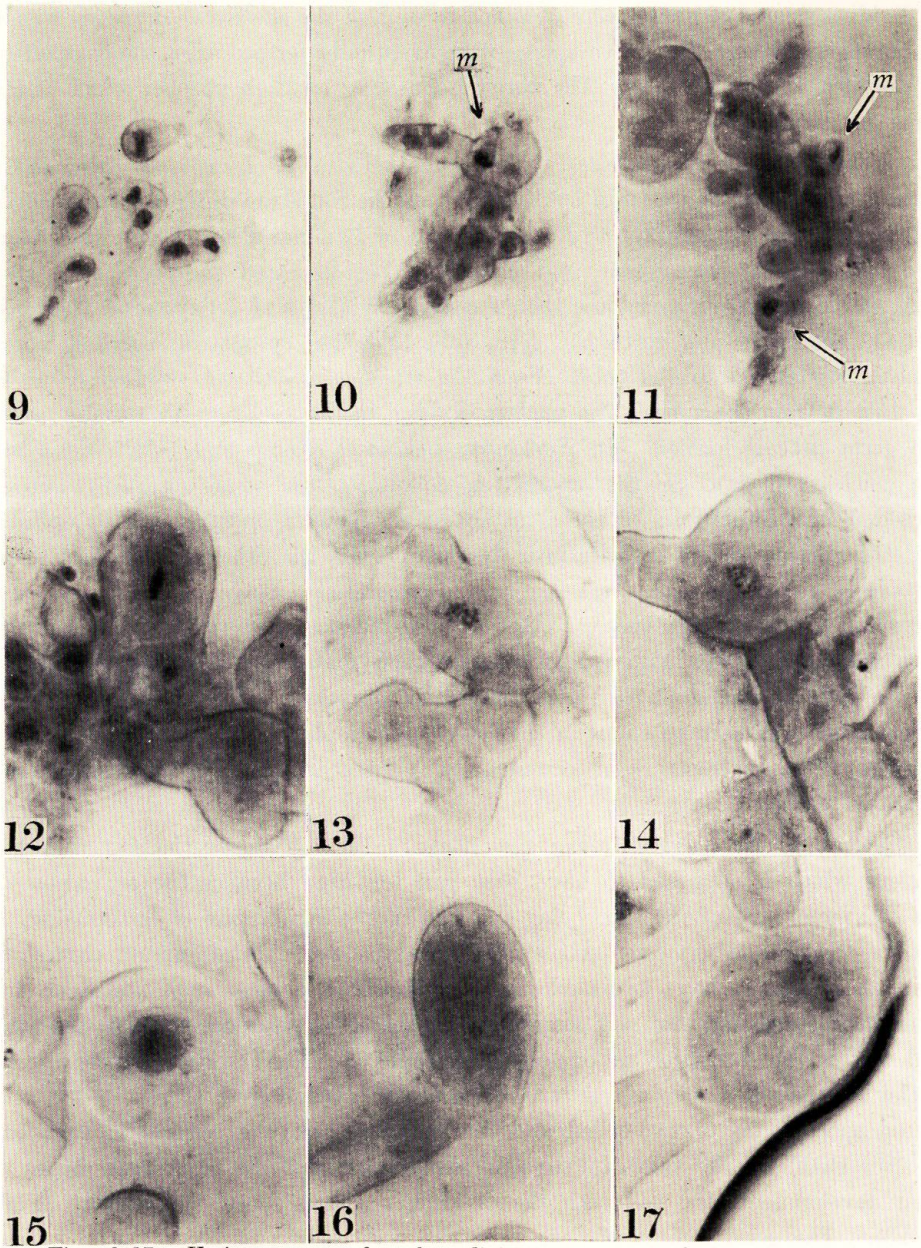
Figs. 1–8. Various stages of nuclear divisions in young sporangia of *Laminaria japonica* Aresch. ×800
1. Synapsis; 2, Drawing of fig. 1; 3 & 5, Early metaphase; 4, Drawing of fig. 3; 6, Side view of metaphase; 7, Side view of metaphase in the second nuclear division; 8, Metaphase in the third nuclear division.

of dividing figures of nuclei were obtained in each of the successive divisions within a sporangium where thirty-two spores were eventually formed after the completion of the fifth nuclear division. The chromosome was n=22 in number which is the same as Abe's count.[1)]

Mitosis in the gametophytes: The gametophytes of *Laminaria japonica* in culture were observed in detail by Kanda[22)] and he found no difference in essential points from the gametophytes of other species of Laminariales. Later, they were also observed by Kinoshita[23)] and Yabu.[24)] The liberated zoospores soon stop their active movement and loose their flagella. Within 3 to 5 days after they had settled, the spore put forth the germ tube and the cytoplasm including spore nucleus migrated into the tube. Soon the transverse wall was formed so as to separate it from the tube. The nuclear division in the spore occurs usually when the germ tube is formed. In *Nereocystis luetkeana*, Kemp and Cole[17)] described the nuclear division in the spores as follows: "In some instances mitosis preceded wall formation; in these instances one nucleus, most of the cytoplasm, and the chromatophore migrate into the tube, while the other nucleus and some cytoplasm remain in the spore-case and degenerate." Such an event was also observed in the present species. About seven weeks after the start of culture, both male and female gametophytes attain their maturity. The dividing nuclei in the cells of gametophytes were frequently observed. The exact number of chromosome was hard to count in the cell of gametophytes, however chromosomes between 16–22 in numer were occasionally counted in the cells of females and rarely in males.

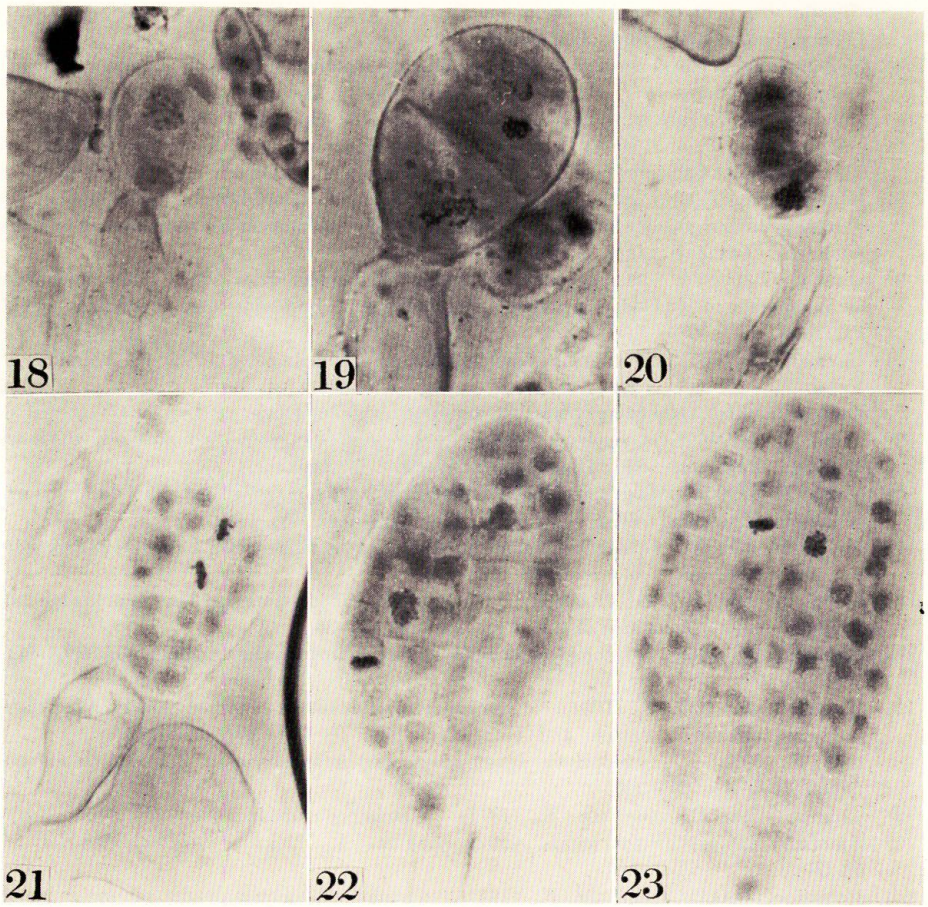
Mitosis in the egg and young sporophytes: Fig. 15 shows the egg with the nucleus which is supposed to have been just fertilized because the formation of the cell membrane of the egg, being the sign of the completion of fertilization, is not yet visible and the egg nucleus filled with the well stained minute chromatin granules is seen to have two nucleoli. The fertilized nucleus soon undergoes the interphase stage and the cell membrane makes its appearence around the egg, and this is the formation of one-celled sporophyte. The dividing figures of the nuclei in one-celled sporophyte was occasionally encountered (Figs. 16–18). In young sporophytes of two-celled and four-celled states, the nucleus in each cell of their thalli usually takes its division simultaneously. In more later stages of their development, the cell with the dividing nuclei is found sporadically (Figs. 21–23). The good figures of late prophase and early metaphase nuclei in the young sporophytes showed to have 30–40 chromosomes, though their number had never been counted exactly.

In conclusion of these results, the chromosome number seen in gametophytes and young sporophytes in culture showed that gametophytes are haplont and sporophytes are diplont.



Figs. 9–17. Various stages of nuclear divisions in gametophytes, eggs and young sporophytes of *Laminaria japonica* Aresch. ×800

9. Nuclei in gametophytes in early stages of their development; 10 & 11. Metaphase (m) in male gametophytes; 12. Side view of metaphase in female gametophyte; 13 & 14. Metaphase in female gametophytes; 15. Nucleus in egg, which is seemed to have been just fertilized; 16. Resting nucleus in one-celled sporophyte; 17. Side view of metaphase in one-celled sporophyte.



Figs. 18–23. Various stages of nuclear divisions in young sporophytes of *Laminaria japonica* Aresch. ×800
18. Telophase in one-celled sporophyte. One of the masses of chromosomes at pole is on the focus; 19. Anaphase in two-celled sporophyte; 20. Metaphase in four-celled sporophyte; 21–23. Young sporophytes with metaphase nuclei.

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