

Faits divers

## Formation of aposporous gametophytes from dedifferentiated young sporophyte cells of *Laminaria japonica* ARESCHOUG (Laminariales, Phaeophyta)\*

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There have been reports on the aposporous formation of male and female gametophytes from the explant tissues in three species of Laminariales. FRIES (1980) reported in *Laminaria digitata* and *L. hyperborea* that the callus was formed from explants of the blade meristematic zone, and the callus cells developed aposporously to male and female gametophytes, which formed sporophytes by fertilization. LEE (1985) reported similar observations in *L. saccharina*.

On the other hand, SAGA *et al.* (1978) reported in *L. angustata* that a single cell from callus-like structure induced in long-term cultivation of the sporophyte directly formed a new sporophyte. FANG *et al.* (1983) and YAN (1984) reported in *Laminaria japonica* and *Undaria pinnatifida* that young sporophytes developed directly from explant tissues. We reported in *Ecklonia cava* (NOTOYA and ARUGA 1989) and *Eisenia bicyclis* (NOTOYA and ARUGA 1990) that callus-like cells developed from the explant and they differentiated directly to new sporophytes.

In this paper we report aposporous formation of gametophytes from dedifferentiated cells of young sporophytes of *Laminaria japonica* under a long period of culture in the laboratory.

A mature sporophyte of *Laminaria japonica* was collected in Ohma, Shimokita Peninsula, Aomori Prefecture, on October 30, 1988. Next day, zoospores released from the sporophyte were collected in laboratory. They were cultured to obtain gametophytes at 15 °C under a

photoperiod of 10L:14D, illuminated by cool white fluorescent lamps at about 20  $\mu$ mol photon  $m^{-2}s^{-1}$  in an incubator (Sanyo M-551).

The gametophytes matured and produced sporophytes by fertilization. When young sporophytes became about 1 cm long, blades were separated from stipes. About 30 pieces of the blade were cultured in a bottle with 100 ml culture medium (PESI) under the same conditions for about one and half a year. The culture medium was changed at three- or four-month intervals.

During the culture, the blade cells were partly dead, whereas the living parts became a globular lump (Fig. 1A). Small and large callus-like cells were observed in the lump when it was lightly squashed on the slide glass by cover slip. When it was squashed more intensively, these cells were separated from each other and became small or large single spherical cells (Fig. 1B). These cells were suspended and cultured in Petri dishes with PESI medium. Germination and filamentous uniseriate growth were observed from each cell after a week (Fig. 1C). The narrow or wide filamentous germlings were grown in culture (Fig. 1D-F), and these filamentous plants matured to be male or female gametophytes in a month (Fig. 1G & H). After another month in culture, the sporophyte developed on the female gametophyte (Fig. 1I).

Thus, the cells of young sporophytes of *Laminaria japonica* were dedifferentiated and became easily separated in long-term culture. The culture system was not axenic, and it is not evident whether the sporophyte cells were separated by bacterial enzymes or not. The separated cells developed aposporously into male and

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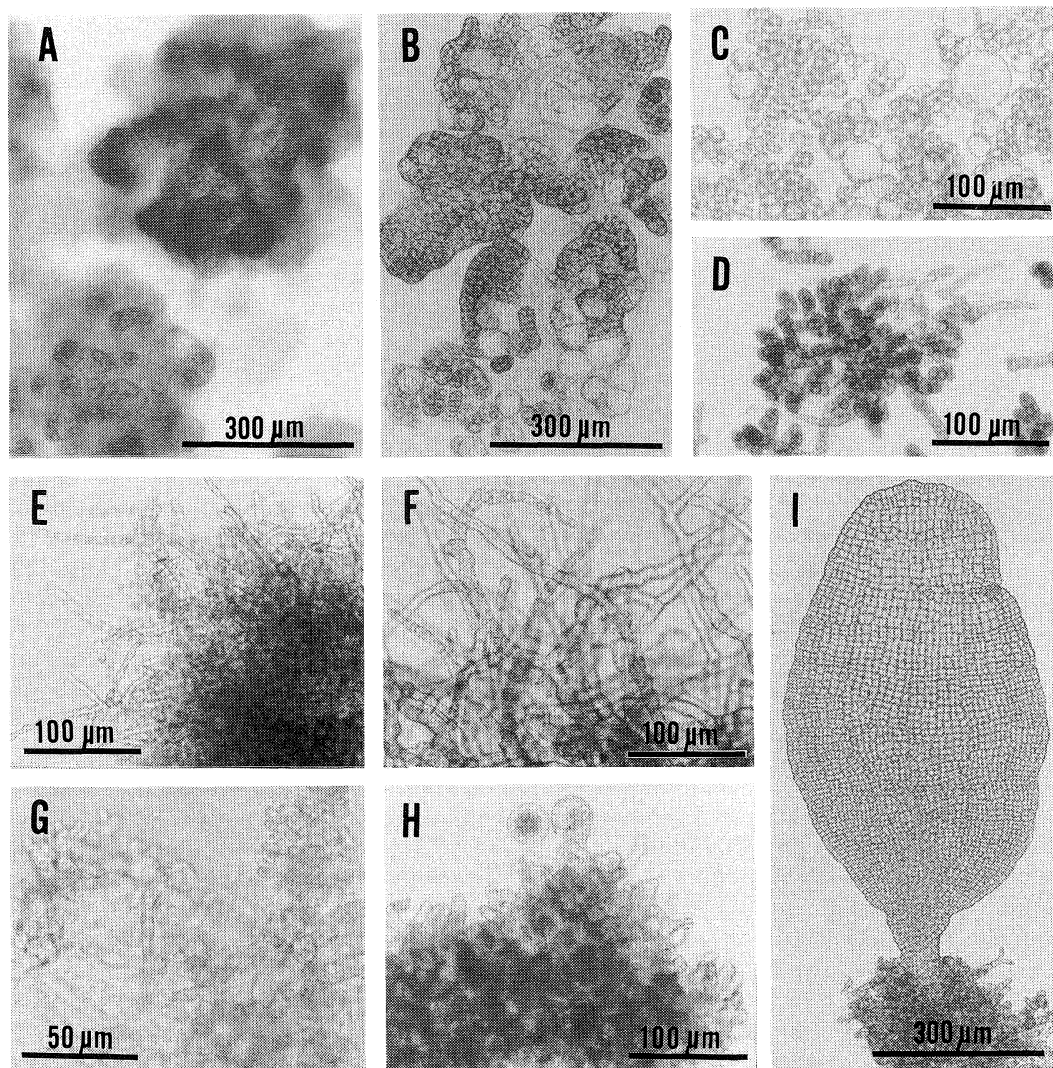


Fig. 1. Formation of aposporous gametophyte from dedifferentiated sporophyte cells of *Laminaria japonica* ARESCHOUG. (A) Globular lump of sporophyte cells in long-term culture. (B) Small and large cells in globular lump after squashed lightly. (C) Separated small and large cells after squashed intensively. (D) Germinated cells suspended in liquid medium after a week in culture. (E) Narrow filamentous germlings likely to be male gametophytes. (F) Broad filamentous germlings likely to be female gametophytes. (G) Narrow filamentous germlings matured and produced spermatial zoospores. (H) Broad filamentous germlings produced eggs. (I) A sporophyte developed on female gametophytes.

female gametophytes, and sporophytes developed on the female gametophytes. Although the cytological evidence was not observed on the developed gametophytes, the results are the same as those reported by NAKAHARA and NAKAMURA (1973) in which isolated single cells from *Alaria*

*classifolia* sporophyte developed into gametophytes. FRIES (1980) and LEE (1985) reported that aposporous gametophytes developed from the tissue of *Laminaria* spp. They found in long-term cultured tissue that aposporous gametophytes developed in the same way as in this

experiment. In *L. angustata*, however, a single cell from callus-like structure induced from young sporophyte in long-term culture developed directly to sporophyte (SAGA *et al.* 1978). Therefore, it seems that aposporous gametophyte formation is induced in the case of nutrient depletion, while in sufficient nutrients is induced direct development of the sporophyte.

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## マコンブ幼胞子体の脱分化細胞からの配偶体形成

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要旨：マコンブの幼胞子体を1年半の長期間にわたって15°C、約20  $\mu\text{mol photon m}^{-2}\text{s}^{-1}$ 、10L:14Dの条件下で培養し続けた結果、胞子体の葉状部が塊状に変化し始めた。この塊状部分をスライドグラス上で軽く押し潰したところ、各細胞は大小の球形細胞となって容易に分離した。分離した細胞を液体培地で培養したところ、それぞれの細胞は発芽して細い糸状または太い糸状の藻体となり、マコンブの雌雄配偶体に似た体に発達した。その後これらの藻体は成熟して卵または精子を形成し、受精が行われて雌配偶体上に胞子体が発達した。