Marine Biology 45, 297-309 (1978)

MARINE BIOLOGY

Light and Temperature Demands for Growth and Reproduction of Laminarian Gametophytes in Southern and Central California

K. Lüning[1,2] and M. Neushul[2]

[1]Biologische Anstalt Helgoland (Meeresstation); Helgoland, Germany (FRG), and
[2]Marine Science Institute, University of California; Santa Barbara, California, USA

Abstract

The gametophytes of 9 laminarian species (4 from southern California, and 5 from central California, USA) became fertile in the unicellular stage (female gametophytes) or in a few-celled stage (male gametophytes), when appropriate temperatures and a sufficiently high quantum irradiance in the blue part of the spectrum were supplied. Vegetative growth, leading to the formation of filamentous gametophytes was light-saturated at relatively low irradiances (4 W m^{-2}; equivalent to about 2 nE cm^{-2} sec^{-1} or an illuminance of 1000 lux), whereas 2 to 3 times this irradiance in continuous fluorescent cool white light was needed to induce the majority of the gametophytes to become fertile. An illuminance of 8300 lux did not inhibit the development of the gametophytes from southern Californian species. *Egregia menziesii* exhibited an exceptionally low quantum demand for induction of fertility. Gametophytes of species from central and southern California differed in regard to their temperature optimum for growth (12°C in the former, 17°C in the latter) and their upper temperature limit for reproduction (17°C in the former, 20°C in the latter).

Introduction

After the discovery of the heteromorphic life history in European genera of the Laminariales by Sauvageau (1915, 1918) the development of the gametophytes was also followed in several laminarian genera occurring along the Pacific coast of North America [see for literature Hollenberg (1939) and Cole (1968)]. Influences of environmental factors, such as temperature, light or nutrients, on the development of laminarian gametophytes have been investigated in Europe by Schreiber (1930), Harries (1932), Kain (1964, 1969), Pérez (1971), Lüning and Dring (1972, 1975), Cosson (1973, 1975, 1977), and Cosson *et al.* (1976), in South Africa by Branch (1974), in Japan by Saito (1956a, b), Segi and Kida (1957), and Yabu (1964), and in China by Tseng *et al.* (1962). On the Pacific coast of North America, similar studies have been conducted only in the case of *Nereocystis luetkeana* (Vadas, 1972) and *Laminaria saccharina* (Hsiao and Druehl, 1971, 1973a,b,c). Anderson and North (1969) determined vegetative growth rates of the gametophytes of *Macrocystis pyrifera* in different spectral ranges.

So many different opinions exist so far regarding the influence of environmental factors on growth and reproduction of laminarian gametophytes (see for literature, Kain, 1964; Hsiao and Druehl, 1971; Lüning and Dring, 1972) that it seems necessary to verify those responses that reflect general traits. The present study represents an approach to determine the specific light and temperature demands for vegetative growth and reproduction of 9 laminarian species from California, some of which are environmentally and economically important plants.

Materials and Methods

Sporogenous plants were sampled at Santa Barbara, southern California, from October until February and from central California in early December. To obtain zoospores, mature sorus pieces were wiped clean and stored overnight in closed crystallizing dishes at 12°C. Zoospores were released in the usual way by immersing a sorus piece for some minutes in enriched seawater (PES; Provasoli, 1968). The zoospores were allowed to

0025-3162/78/0045/0297/$02.60

settle on cover glasses (18 x 18 mm) at a density of 600 to 1000 spores per cover glass. The cover glasses with the adhering spores were left in darkness for 24 h and then placed into small Petri dishes and cultivated at constant temperature (± 0.5 C°) in incubators at different distances from the overhead light source. To eliminate diatoms, an additive of germanium dioxide (1 ml of an aqueous saturated solution of GeO_2 per liter of PES) was used for the first 4 days. PES was changed every week from then onwards.

The light in each incubator was provided by 2 cool white fluorescent lamps (General Electric F15T12/CW/15 Watt). Continuous light was used in the incubators. The quantum irradiance was measured by means of a Lambda quanta meter (Lambda Instruments Corporation, Lincoln, Nebraska). The photometric equivalent for 1 nE cm^{-2} sec^{-1} of the white fluorescent light was 520 lux, the energetic equivalent about 2 W m^{-2}. Using the quanta meter in combination with the Schott cut-off glass filter GG 495, it was determined that 20% of the quantum irradiance in the white light incubator fields was within the range 400 to 500 nm. Spectral light fields were obtained by combining 24 V 250 W quartz-iodine lamps in Leitz Prado Universal projectors with Schott glass filters. BG 12 (transmitted waveband <400 to 530 nm) was used in the blue, the glass filter combination SFK 11 (transmitted waveband 525 to 570 nm) in the green, and the glass filter combination SFK 15 (transmitted waveband 610 to >700 nm) in the red. Red fluorescent lamps combined with red Plexiglas were used as broad-band red field (no light transmitted at wavelengths <600 nm as measured with an Isco Spectroradiometer).

For permanent preservation, the gametophytes were fixed on the cover glasses in 4% formalin for 2 h, briefly washed in distilled water and then stained in a 5% aqueous alizarine viridine solution for several hours (stock solution: 1 g of alizarine viridine, obtained from Chroma-Gesellschaft, Stuttgart-Untertürkheim, FRG, dissolved in boiling 5% potassium-chromium-sulfate solution). The cover glasses with the adhering gametophytes were again briefly washed in distilled water and then embedded in 15% glycerine, diluted with distilled water. After concentration of the glycerine, this procedure gave a shrinkage rate of

Table 1. Origin of samples and geographical distribution of species investigated. LAM: Laminariaceae; ALA: Alariaceae; LES: Lessoniaceae

Family and species		Sample locality	Geographical distribution	Vertical distribution[b]
Species occurring in Southern California				
LES	*Macrocystis pyrifera* (L.) C.Ag.[a]	Santa Barbara (34°23')	Alaska to Baja California, South America, South Africa, Southern Australia[b]	Subtidal
LAM	*Laminaria farlowii* Setch.	Santa Barbara (34°23')	Central to southern California (isolated collection at Comox, British Columbia)[c]	Low intertidal (N. California) to subtidal
ALA	*Pterygophora californica* Rupr.	Santa Barbara (34°23')	British Columbia to Baja California[b]	Subtidal (occasionally low intertidal)
ALA	*Egregia menziesii* (Turn.) Aresch.	Santa Barbara (34°23')	Alaska to Baja California[b]	Midtidal to subtidal
Species occurring in Central California				
LES	*Macrocystis integrifolia* Bory	Pillar Point (37°31')	British Columbia to Central California; western coast of South America[b]	Lower intertidal to shallow subtidal
LAM	*Hedophyllum sessile* (C.Ag.) Setch.	Bodega Bay (38°16')	Alaska to Monterey County[b]	Midtidal to subtidal
ALA	*Alaria marginata*	Pillar Point (37°31')	Alaska to Monterey County[b]	(Low intertidal)
LAM	*Laminaria dentigera* Kjellm.	Bodega Bay (38°16')	Bering Strait to Baja California[b]	Lower intertidal to upper subtidal
LAM	*Laminaria sinclairii* (Harv.) Farl., Anders & Eaton	Pillar Point (37°31')	British Columbia to Ventura County, California[c]	Low intertidal

[a] As noted in Abbott and Hollenberg (1976), there is some question as to the specific identity of the *Macrocystis* population at Santa Barbara, which has provisionally been called *M. angustifolia* (Neushul, 1971).

[b] According to Abbott and Hollenberg (1976).

[c] According to Druehl (1968).

the cells of about 10% compared to live cells. Vegetative growth of young gametophytes was determined in the preserved material by measuring 20 female gametophytes by means of a micrometer eye piece. The percentage of fertile gametophytes was determined in the live cultures by counting 250 plants in groups of 50. Least significant ranges were calculated according to Sokal and Rohlf (1969).

Results

In all species investigated (Table 1), the diameter of the embryospore, after 3 days cultivation in darkness, was 4 to 5 µm. Under optimum conditions of quantum irradiance (4 to 6 nE cm^{-2} sec^{-1}) and temperature (12° to 17°C), this diameter increased by 3 to 5 times after 1 week of growth, in continuous fluorescent cool white light (Figs. 1-3). At this time many of the unicellular female gametophytes had started to elongate to form oogonia or another vegetative cell, or had formed as many as 2 to 4 cells. Under the same quantum irradiance conditions and temperature, the male gametophytes consisted of 5 to 10 cells after 1 week. The increase in diameter of the primary cell of the female gametophyte is a good parameter for estimating

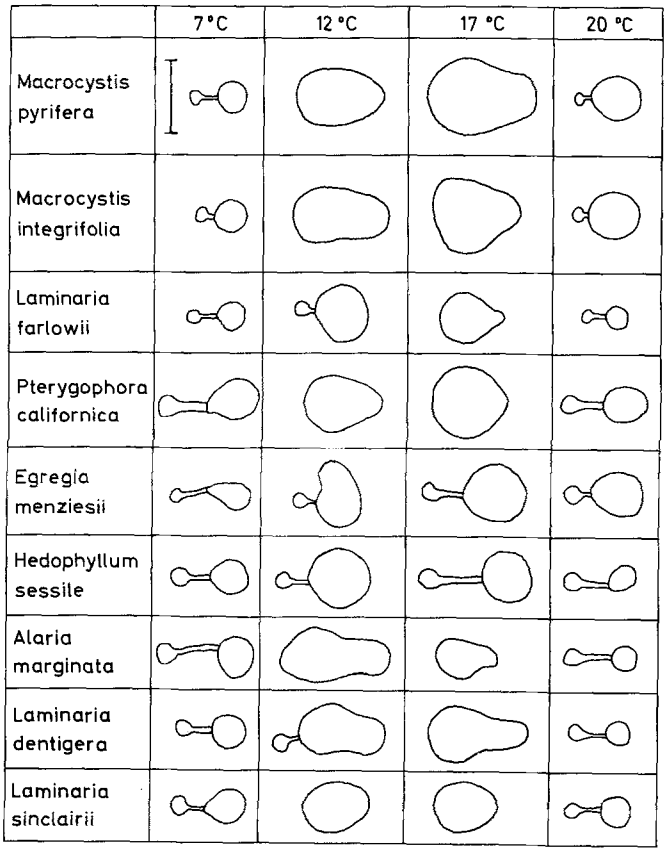

Fig. 1. Californian Laminariales. Records of the most advanced unicellular, female gametophytes at age of 1 week, cultivated at a quantum irradiance of 4 nE cm^{-2} sec^{-1} (cool white fluorescent light) and at 4 temperatures. Scale bar = 20 µm

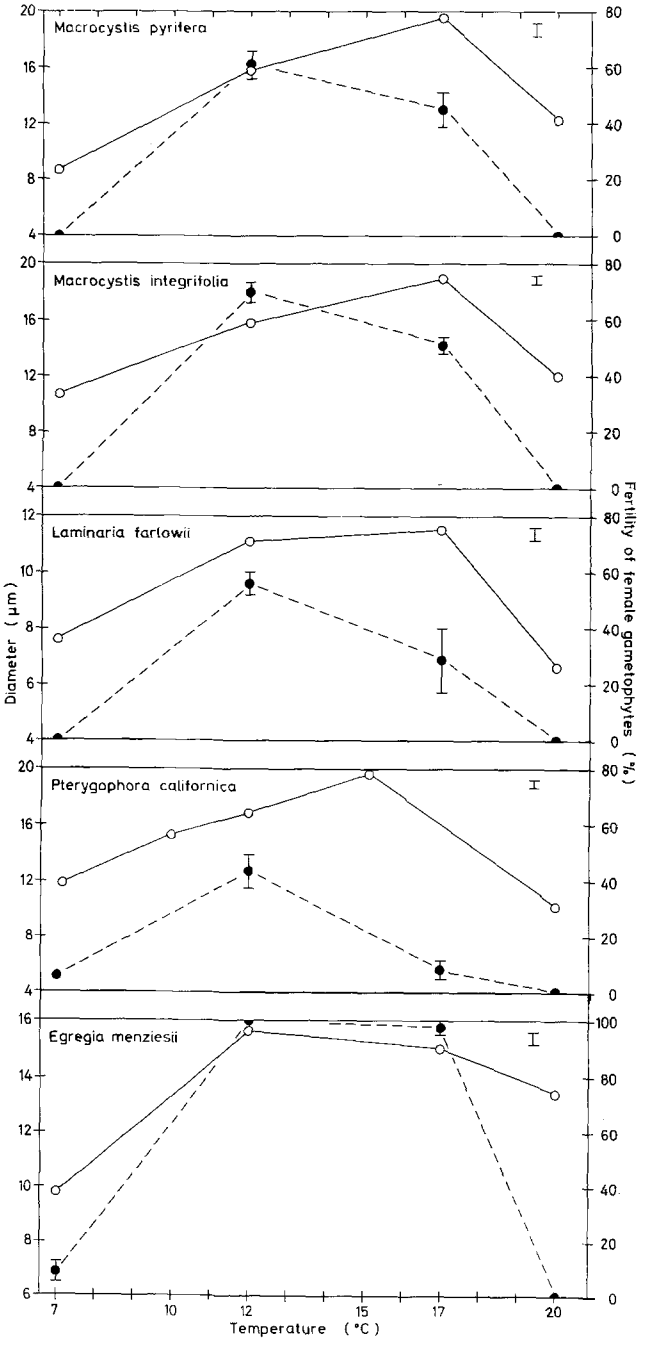

Fig. 2. Laminarian species from southern California and *Macrocystis integrifolia*. Open circles: diameter of primary cell of female gametophytes at age of 1 week (n = 20; vertical bars in right corners indicate least significant ranges at 5%); filled circles: percentage of fertility of female gametophytes at age of 2 weeks (n = 250; vertical bars indicate 95% confidence limits). All plants cultivated in continuous light at 6 nE cm-2 sec-1

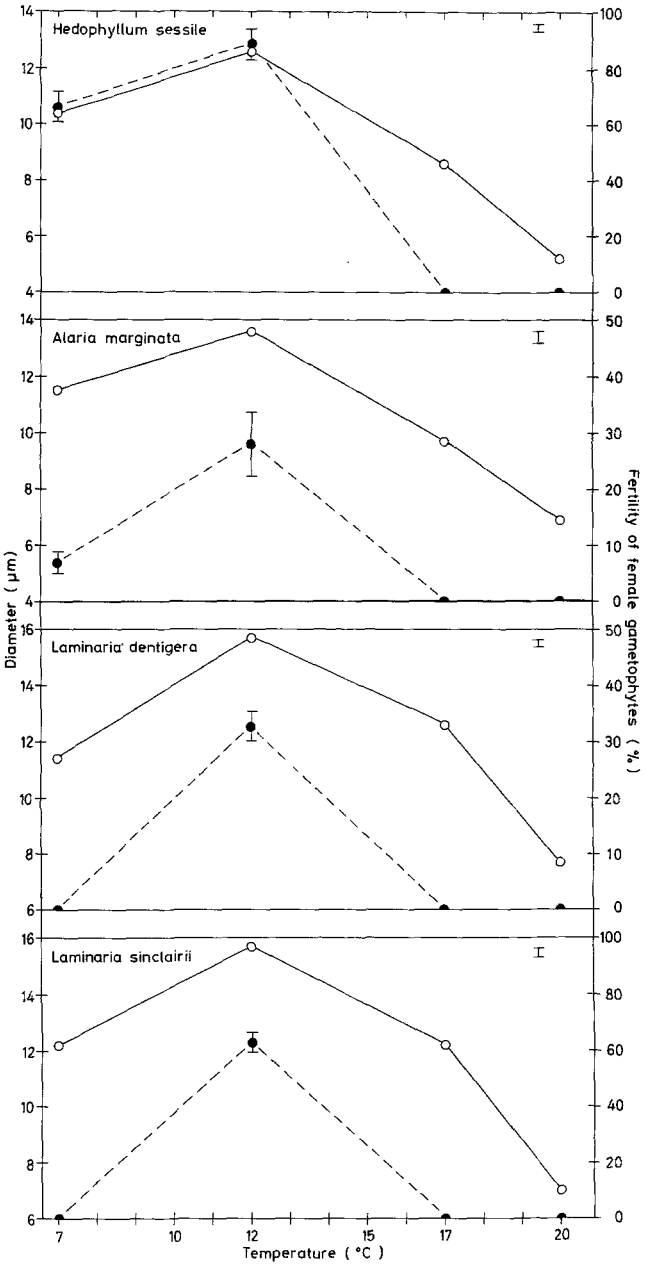

Fig. 3. Species from central California. For further details see legend to Fig. 2

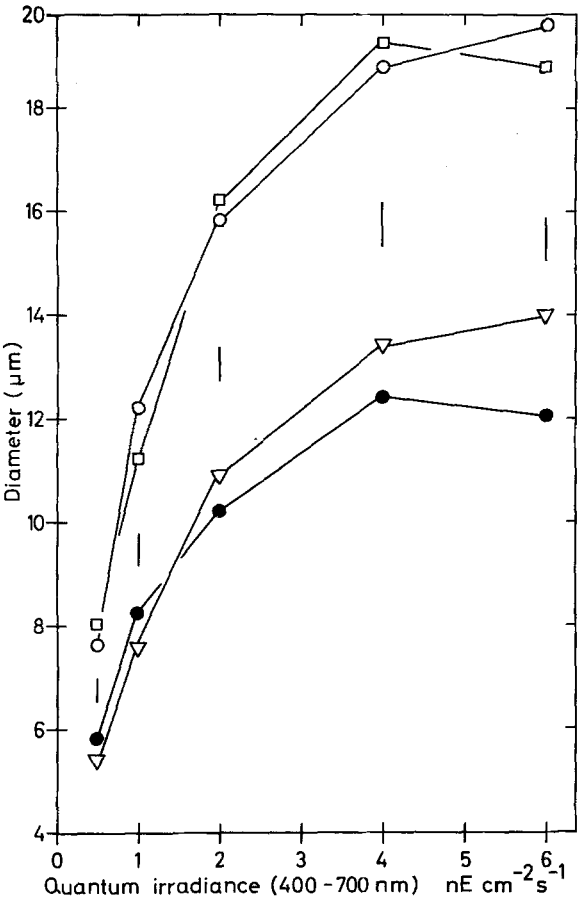

Fig. 4. *Macrocystis pyrifera* (open circles), *M. integrifolia* (squares), *Laminaria farlowii* (filled circles), and *Egregia menziesii* (triangles), cultivated at 17°C for 1 week. Diameter of primary cell of female gametophyte at different quantum irradiances in continuous cool white fluorescent light. Vertical bars: least significant ranges at 5% for each quantum irradiance

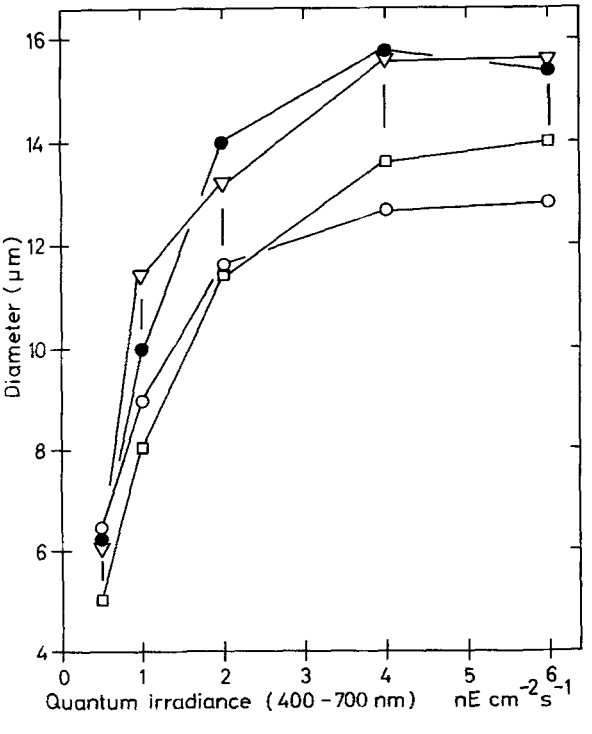

Fig. 5. *Hedophyllum sessile* (open circles), *Alaria marginata* (squares), *Laminaria dentigera* (filled circles), and *L. sinclairii* (triangles), cultivated at 12°C. For further details see legend to Fig. 4

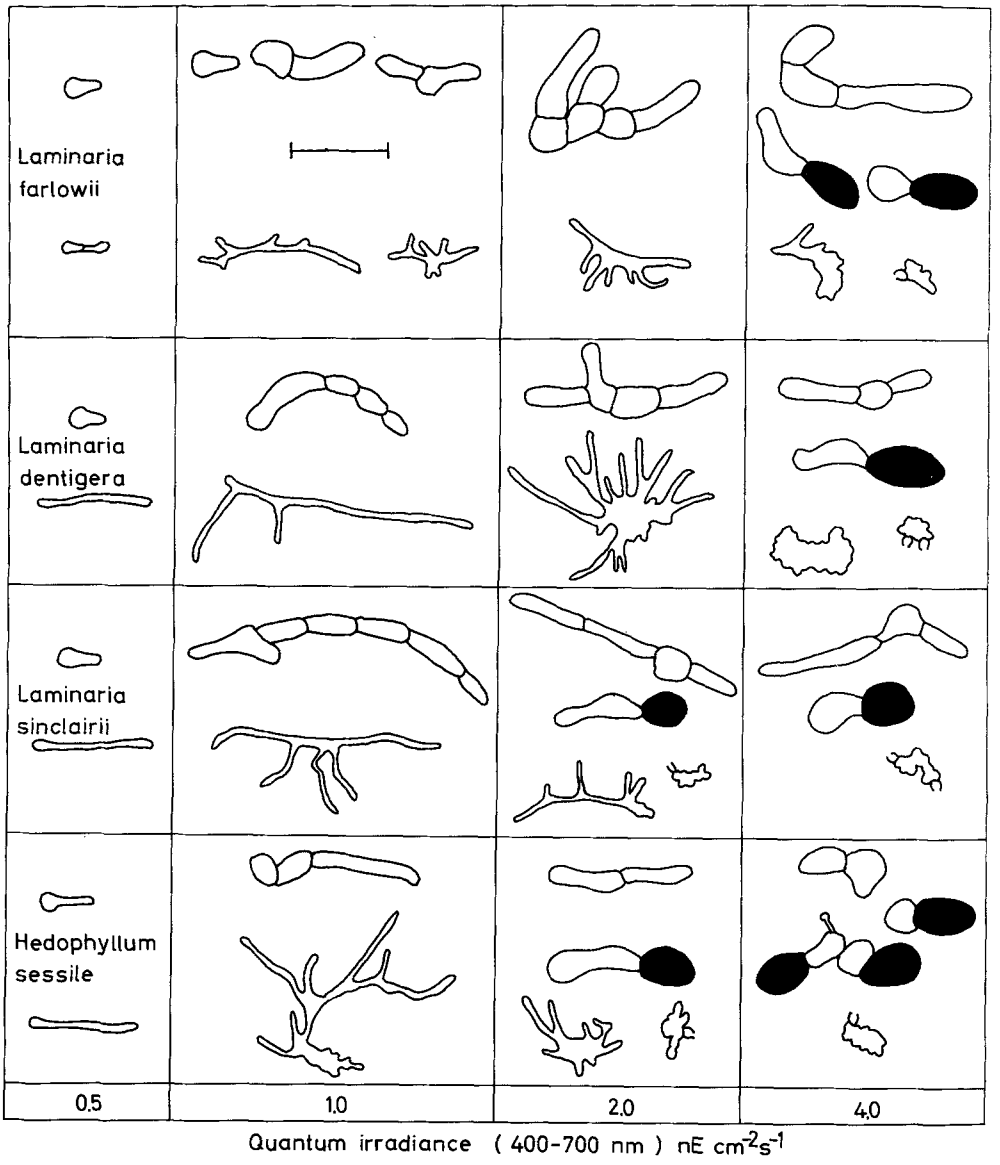

Fig. 6. Gametophytes of Laminariaceae. Records of most advanced vegetative and fertile plants, cultivated at 12°C and at 4 quantum irradiances in continuous cool white fluorescent light for 2 weeks. For each condition, vegetative female gametophytes are shown above, fertile female gametophytes in the middle (if fertility >10%; eggs and zygotes filled-in), and male gametophytes below (only outlines of the multicellular males are given; U-shaped portions represent emptied antheridia, if fertility >10%). Scale bar = 50 µm

growth rate, since once the female gametophyte becomes multicellular it is difficult to follow its growth quantitatively because of the irregular shape of the cells and because of its multidirectional branching.

The influence of temperature on the vegetative growth of the southern and central Californian species was different. Northern species (Fig. 3) had a growth optimum at 12°C and grew better at 7°C than at 20°C, while southern species (Fig. 2) had their growth optimum at 17°C and grew better at 20°C than at 7°C (*Macrocystis pyrifera, Egregia menziesii*) or only slightly better at 7°C than at 20°C (*Laminaria farlowii, Pterygophora californica*). *E. menziesii* grew as well at 17°C as at 12°C. Another exception was provided by *M. integrifolia*, which showed almost the same pattern of temperature response as *M. pyrifera*.

The influence of quantum irradiance on vegetative growth is shown in Figs. 4 and 5. Vegetative growth is light-saturated from about 2 nE cm^{-2} sec^{-1} onwards. Between 0.5 and 1.0 nE cm^{-2} sec^{-1}, vegetative growth and probably photosyn-

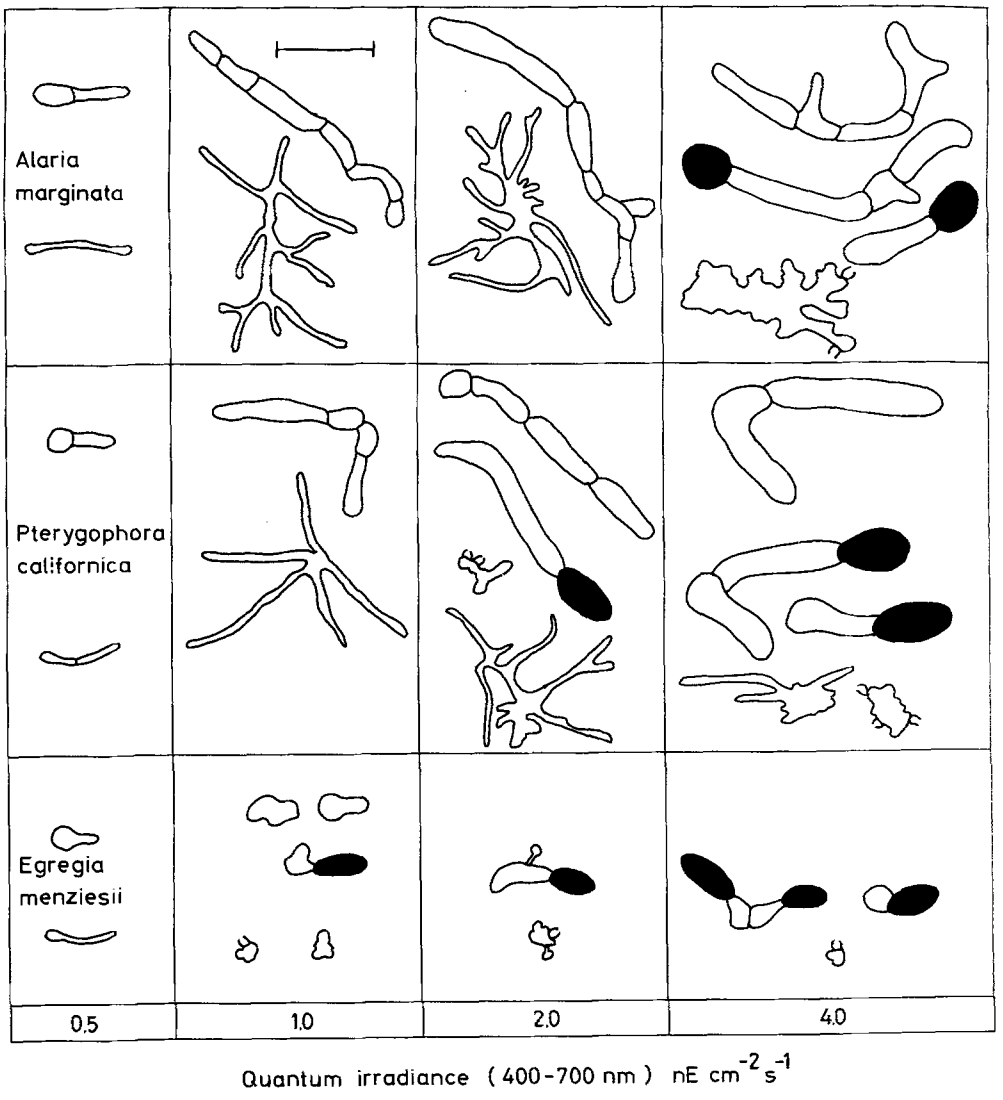

Fig. 7. Gametophytes of Alariaceae. For further details see legend to Fig. 6

thesis of the female gametophytes increases linearly with quantum irradiance (Figs. 4 and 5). However, the process of cell division obviously does not depend on attaining maximal cell diameter (14 to 21 µm at 4 nE cm^{-2} sec^{-1}; Fig. 1). Cell division occurs at values as low as 0.5 nE cm^{-2} sec^{-1}, when the diameter of the primary cell of the female gametophyte is only 8 to 12 µm (Figs. 6-8). At this low quantum irradiance (and at 17°C), the percentages of female gametophytes which had produced a second cell after 2 weeks were: *Macrocystis pyrifera*, 46%; *Laminaria farlowii*, 82%; *Pterygophora californica*, 96% (in each n = 200). Assuming a linear relationship between quantum irradiance and the percentage of two-celled female gametophytes produced, then a total number of quanta (400 to 700 nm; cool fluorescent white light) of 320 to 660 µE cm^{-2} would allow 50% of the female gametophytes to divide and then form multicellular plants (Fig. 11). The male gametophytes of all species investigated had formed 3 to 5 cells at 0.5 nE cm^{-2} sec^{-1} (12° and 17°C) at an age of 2 weeks, so their quanta demand for growth and subsequent division is even lower than in the female gametophytes. At 1.0 nE cm^{-2} sec^{-1}, at an age of 2 weeks, the filamentous gametophytes which had not been induced to become fertile were nearly about the same in cell number and size as filamentous gametophytes at higher irradiances (Figs. 6-8). This finding indicates again that saturation of vegetative growth, and probably of photosynthesis, takes place between 1 and 2 nE cm^{-2} sec^{-1}.

A quantum irradiance as high as 16 nE cm^{-2} sec^{-1} (cool fluorescent white

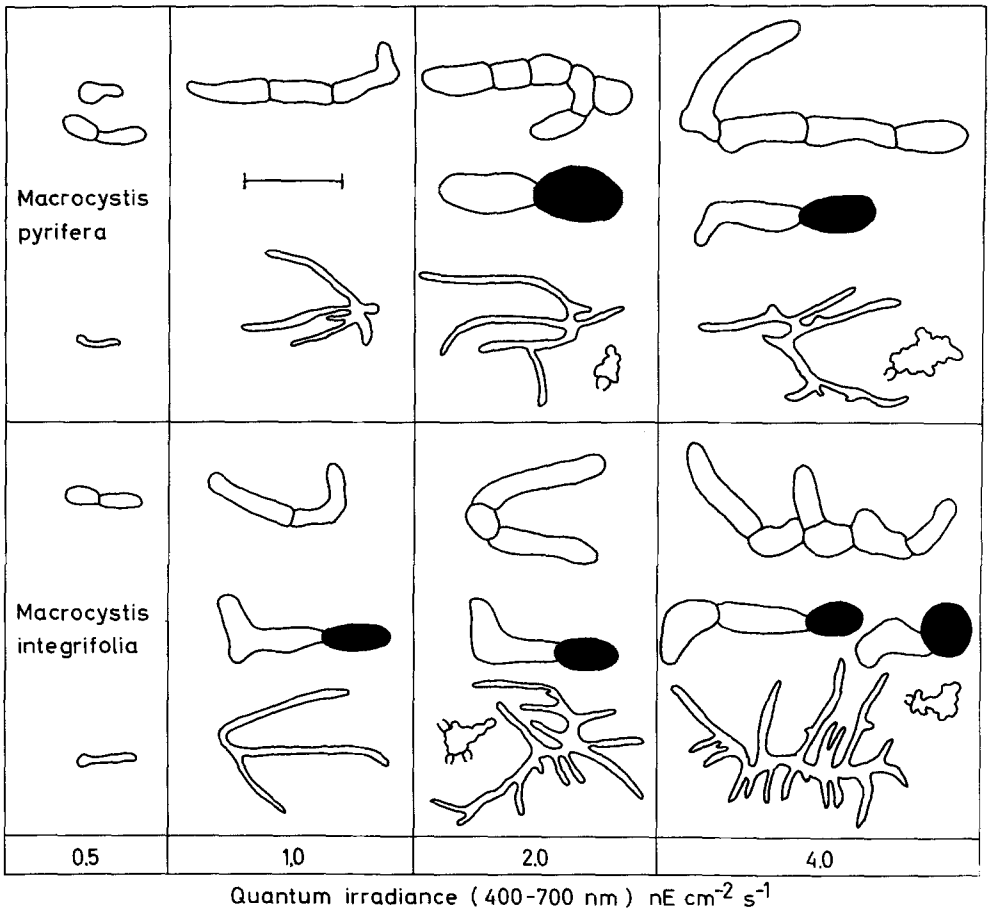

Fig. 8. Gametophytes of Lessoniaceae. For further details see legend to Fig. 6

light) – corresponding to an illuminance of 8300 lux – did not inhibit development of the southern Californian species. Direct sunlight, however, killed the spores after some minutes. One-day old embryospores of *Macrocystis pyrifera, Laminaria farlowii* and *Pterygophora californica* survived only 1 to 4 min of direct sunlight (90 to 110 nE cm^{-2} sec^{-1}; open sky in December; spores covered by 1 cm of enriched seawater; water temperature 12° to 18°C) but not 8 min. *Egregia menziesii* survived 8 min exposure in the same experiment, but not 15, 30 and 60 min.

To obtain high fertility in female or male gametophytes after 2 weeks of cultivation in white fluorescent light, much higher quantum irradiances are necessary than for vegetative growth. Several species, e.g. *Macrocystis pyrifera,* exhibited an almost linear dependence on quantum irradiance up to 6 nE cm^{-2} sec^{-1} (Fig.9). Fertility after 3 to 4 weeks was higher also at lower irradiances, (down to 1 nE cm^{-2} sec^{-1}). At 0.5 nE cm^{-2} sec^{-1}, however, gametophytes did not mature even after 4 weeks. *Egregia menziesii* was by far the fastest species to mature. In fact, 59% of its female gametophytes had released eggs already at an age of 8 days at 6 nE cm^{-2} sec^{-1} (12°C).

Lower temperatures favored the onset of fertility in the gametophytes of all species investigated (Figs. 2 and 3). The optimum temperature was, among those used, 12°C. The upper temperature limit for reproduction was different in the central and southern Californian species. While no reproduction occurred at 17°C after 2 weeks in the former group, the reproduction of the latter was only inhibited at 20°C. *Macrocystis integrifolia* behaved again in this regard like *M. pyrifera*. A temperature of 7°C is probably not totally inhibitory for the reproduction of the species from southern California, since it was found in another experiment that some gametophytes of *Macrocystis pyrifera* became fertile at this temperature within 3 weeks.

Most female gametophytes became fertile in the unicellular stage (Figs. 6-8). Female gametophytes which had not been induced to become fertile had formed 2 to 6 elongated cells at quantum irradiances ranging from 1 to 6 nE cm^{-2} sec^{-1}

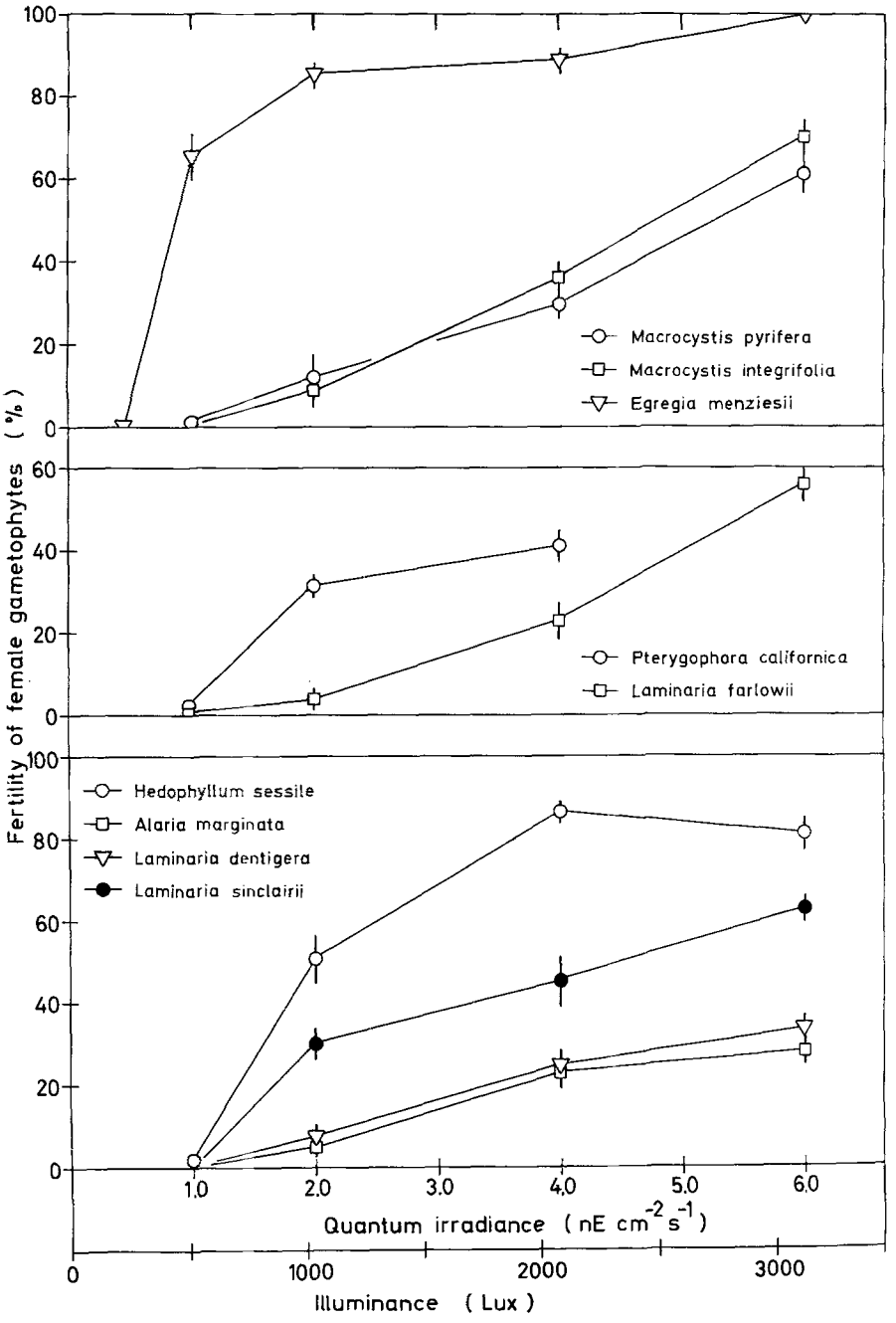

Fig. 9. Californian Laminariales. Percentage of fertile female gametophytes at age of 2 weeks, cultivated at 12°C and at 4 quantum irradiances/illuminances) in continuous cool white fluorescent light. Vertical bars = 95% confidence limits

at an age of 2 weeks (12° and 17°C). Also, vegetative male gametophytes exhibited filamentous growth, whereas fertile males consisted of almost isodiametric cells and had a compact appearance. Regarding the percentage of fertile plants, the reaction of the male gametophytes did not obviously differ from the reaction of the females under different conditions of quantum irradiance and temperature although the behaviour of the males was not followed in detail.

The influence of light quality was investigated by cultivating the southern Californian species for 18 days in continuous blue, green and red light at 6 nE cm^{-2} sec^{-1} and 12°C. Reproduction of female and male gametophytes occurred only in blue light, not in green and red light in *Macrocystis pyrifera*, *Laminaria farlowii* and *Pterygophora californica* (Table 2).

Table 2. Percentages of fertile female gametophytes of laminarian species at age of 18 days in blue light (BG 12), green light (SFK 11) and red light (SFK 15), at 6 nE cm^{-2} sec^{-1} and 12°C. Each value is based on 200 plants

Species	Blue	Green	Red
Macrocystis pyrifera	94	0	0
Laminaria farlowii	100	0	0
Pterygophora californica	100	0	0
Egregia menziesii	100	81	36

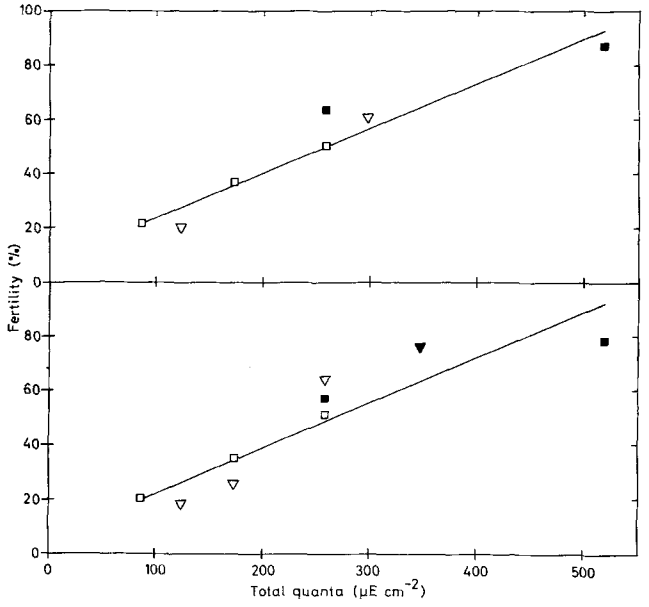

Fig. 10. *Macrocystis pyrifera* (upper graph) and *Pterygophora californica* (lower graph). Fertility of female gametophytes cultivated in red light at 14°C and induced by 1 to 2 nE cm^{-2} sec^{-1} of blue light (BG 12) for 24 h (open squares), 41 h (open triangles), 48 h (filled triangles), or 72 h (filled squares). Values are based on counts of 250 plants. Regression lines have equations: $y = 0.16 x + 7.35$ (upper), and $y = 0.17 x + 5.25$ (lower)

In another experiment, which had a duration of 4 weeks, 6% of the female gametophytes of *M. pyrifera* and *P. californica* became fertile also in red light (2 nE cm^{-2} sec^{-1}). *Egregia menziesii* again provided an exception, since it exhibited considerable fertility also in green and red light (Table 2).

About 260 µE cm^{-2} in the blue range are required to induce 50% of the female gametophytes of *Macrocystis pyrifera* and *Pterygophora californica* to become fertile at 14°C (Fig. 10). This result was achieved by growing the gametophytes from zoospores in red fluorescent light at 2 nE cm^{-2} sec^{-1} (14°C) and irradiating the plants (females with 1 to 3 cells) at an age of 18 days for 12 to 72 h under 1 or 2 nE cm^{-2} sec^{-1} of blue light (Leitz Prado Universal projector with Schott glass filter BG 12). The gametophytes were then returned to the red light field and the percentage of fertile female gametophytes was determined 10 days after the beginning of the blue light treatment.

Discussion

It is of interest to distinguish those features of laminarian gametophytes that are of general occurrence from those that are found in only certain species. Obviously, there are specific differences regarding the optimum temperature for vegetative growth as well as for the upper lethal temperature limit of the gametophyte (Kain, 1969; Branch, 1974). Also, the upper temperature at which reproduction stops but vegetative growth still proceeds seems to differ according to the latitudinal range which a given species inhabits (Figs. 2 and 3). One may go even a step further and predict that species which occur over a wide range of latitudes, with different temperature climates, have not only developed into different morphological forms [e.g. *Egregia menziesii*: forms differing in stipe and sporophyll morphology (Abbott and Hollenberg, 1976), *Macrocystis pyrifera*: form near Santa Barbara, described provisionally as *M. angustifolia* because of differences in holdfast morphology (Neushul, 1971)], but also into different physiological ecotypes adapted to colder or warmer temperature ranges by changes in the concentrations of certain enzymes or by introduction of isoenzymes with different temperature dependences (Larcher, 1975).

Regarding the influences of irradiance on photosynthesis and vegetative growth of laminarian gametophytes, these can be characterized as extreme-shade plants, since light saturation of photosynthesis [at about 2 to 4 W m^{-2} in *Laminaria hyperborea* (Kain, 1964; her Fig. 7b)] and of vegetative growth [(at about 4 W m^{-2} in the Californian species, according to Figs. 4-5 of present paper; at about 1 to 2 W m^{-2} in *L. digitata* (Cosson, 1975)] occurs at even lower irradiances than in deep-growing red algae, which are photosynthetically light-saturated

at about 11 W m^{-2} (Mathieson and Norall, 1975).

In contrast to vegetative growth, reproduction of the laminarian gametophytes requires (at temperatures above 10°C) a rather high amount of blue quanta, 260 µE cm^{-2} for a 50% induction, in the presently investigated southern Californian species (except in *Egregia menziesii*); this is similarly high in *Laminaria saccharina* (200 µE cm^{-2}; Lüning and Dring, 1975). This requirement of blue light will neither be fulfilled in coastal turbid water with predominantly green light at greater depths, nor in the laboratory in artificial light fields whenever a too small amount of blue light is offered to the plants (tungsten lamps, warm white fluorescent lamps, too low white light irradiances in general). For continuous cool fluorescent white lamps which emit 20% of their total visible quanta in the blue part of the spectrum, an attempt is made to demonstrate the dependence of the morphological development of gametophytes on quantum irradiance (Fig. 11). Since it must be expected that the gametophytes need several days' development from the stage of the embryospore onwards before they react optimally to blue light, it has been – rather arbitrarily – assumed in the present example that this reaction occurs from an age of 1 week onwards if the plants are cultivated in continuous white light. At 0.5 nE cm^{-2} sec^{-1} (260 lux in the case of the presently used lamps), after 1 week 50% of the female gametophytes have received enough quanta to become two-celled. It would take, however, about 5 further weeks for 50% of them to become fertile, provided they do not need more quanta in the multicellular stage for this purpose than if they were one-celled. In fact, Kain (1964) found a few fertile gametophytes in *L. hyperborea* after some months in cultures grown on a 12 h light:12 h dark cycle in cool fluorescent white light at an irradiance as low as 0.22 W m^{-2} (0.1 nE cm^{-2} sec^{-1}).

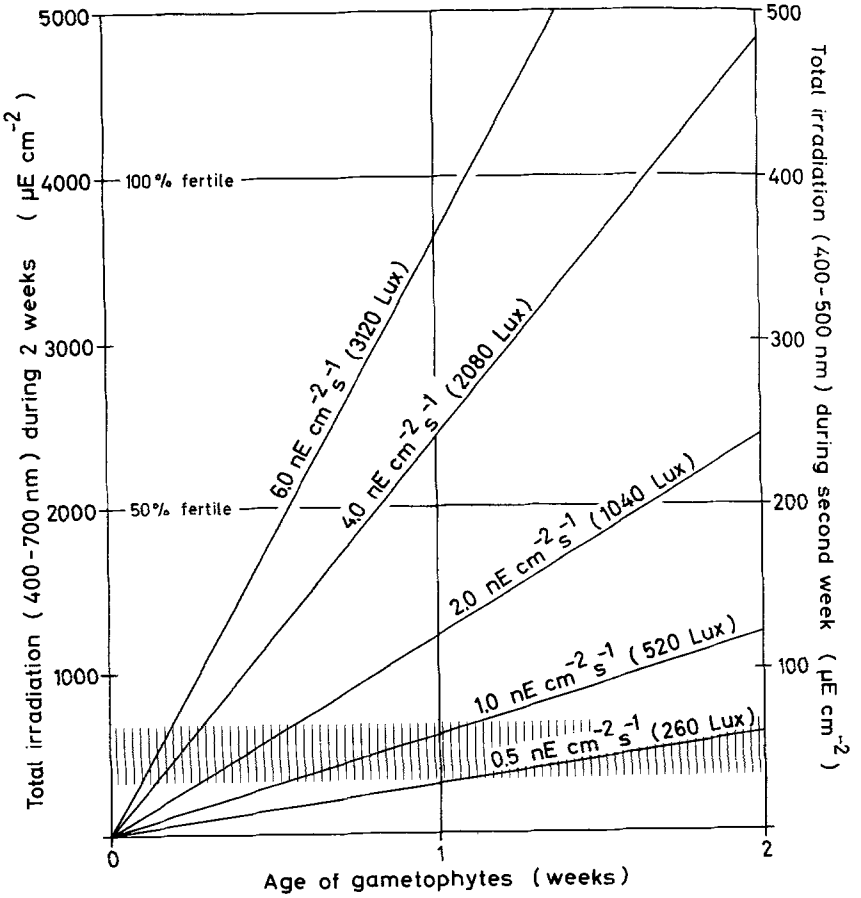

Fig. 11. Total quantum irradiation in the visible range (400 to 700 nm; during 2 weeks) and in the blue range (400 to 500 nm; during the second week) in continuous cool white fluorescent light at different quantum irradiances or illuminances. Hatched area indicates range of total quantum irradiation (400 to 700 nm) allowing vegetative cell divisions. 200 µE cm^{-2} of blue light (400 to 500 nm), received during the second week, have been assumed to induce 50% of the gametophytes to become fertile. See text for further explanation

Fig. 11 shows furthermore that also in cultures grown at 2 nE cm^{-2} sec^{-1} (1040 lux), about half the female gametophytes would be found in the filamentous stage after 2 weeks, while the other half would become fertile mostly in the unicellular stage. Only higher quantum irradiances (4 to 6 nE cm^{-2} sec^{-1}, equivalent to 2080 to 3120 lux) induce most of the female (and male) gametophytes to become fertile due to the number of blue quanta which they received during the second week in continuous white light.

Unfortunately, the basic mechanism in blue-light-mediated photomorphogenesis, which is involved in the induction of fertility in laminarian gametophytes (Lüning and Dring, 1972, 1975), is not well understood at present (Mohr, 1972; Smith, 1975). It is, therefore, hard to interpret all findings which have been made so far on the influence of light quality on the gametophytes of the Laminariales. There is clearly a temperature effect involved in induction of fertility, since at lower temperatures its onset is facilitated (Schreiber, 1930; Kemp and Cole, 1961; Kain, 1964; Pérez, 1971; Branch, 1974) and fertile gametophytes can be obtained also in red light at 10° and 5°C (Lüning and Dring, 1972). The finding that *Egregia menziesii* at 12°C becomes fertile in red and green light (although at a smaller percentage than in blue light; Table 2) might indicate that the temperature range where this is possible also in other laminarian gametophytes has been shifted upwards in this species. Thus, it appears as if the block which prevents reproductive development, and which has to be removed during epigenesis of the primary cell of the female gametophyte and also in the few-celled male gametophyte, can be overcome at lower temperature by an appropriate number of quanta of any spectral range, at medium temperatures only by a sufficient number of blue quanta, while at high temperatures, near the upper lethal limit, where vegetative growth still proceeds, reproduction finally is blocked totally.

One might expect other results than those reported here if one changed the nutrient conditions. However, Hsiao and Druehl (1973a) showed that 588 µg-at NO_3-N/l (per litre) and 15 µg-at PO_4-P/l (PES: 555 µg-at NO_3-N/l and 31 µg-at PO_4-P/l) were optimal for germination of the embryospore, active division and high percentage fertility. So one would not expect differential effects of N and P at relatively high concentrations on vegetative growth and reproduction. Harries (1932) found that insufficient nitrate concentration enhanced vegetative growth instead of percentage fertility, but she used tungsten light, and so one would expect that she worked at limiting conditions of blue light supply which makes it difficult to sort out the differential effects of certain ions and light effects.

It should be emphasized that vegetative and reproductive development represent antagonistic pathways, and vegetative growth stops when all cells of a gametophyte are induced to become fertile. Indeed, one can postulate that environmental conditions are not optimal for inducing reproductive development if the female gametophyte consists of more than one cell before it becomes fertile. Since vegetative growth is almost light-saturated at an illuminance of 1000 lux in white light (Figs. 4 and 5), earlier observers, impressed by the "good development" of the plants, often maintained low illuminances when cultivating gametophytes, not realizing that induction of fertility required twice this illuminance or more in continuous light. Inserting a daily dark period instead of using continuous light decreases – especially at lower illuminances – the chance of the gametophytes becoming fertile. At the time when the critical dose of 200 to 260 µE cm^{-2} of blue quanta for a 50% induction has been supplied, the gametophytes will consist of many more cells than under continuous illumination, since the plants have simply had more time to divide. It is still an open question if more blue quanta are needed for induction of fertility in filamentous than in unicellular female gametophytes. Evidence that it may be so was already provided by Hollenberg (1939) who wrote: "When gametophytes of *Eisenia arborea* in the cultures once take on an elongate sterile form, I have been unable to induce the formation of sex organs, although I have tried low temperatures (2-3°C) and changes in light intensity as well as changes in nutrient concentrations."

It is also still necessary to determine if genuine photoperiodic responses are involved in the development of the species studied. No photoperiodism has been found in the case of *Laminaria saccharina* (Lüning and Dring, 1975), or *L. digitata* (Cosson, 1977). Decreasing percentages of fertile gametophytes with increasing length of the daily dark period has been reported by Hsiao and Druehl (1971; maximum illuminance used, 1075 lux) in the case of *L. saccharina*, and by Vadas (1972; maximum illuminance used, about 2000 lux) in the case of *Nereocystis luetkeana*. The rather low illuminance and the insertion of daily dark periods may

partly explain why Hsiao and Druehl (1971) only obtained filamentous gametophytes of *L. saccharina* in the laboratory, whereas they observed fertile, unicellular female gametophytes of this species in the field (Hsiao and Druehl, 1973c). The same combination of environmental factors and also the use of too high temperatures may also explain why earlier investigators observed in the laboratory maturation only after many weeks in gametophytes which were filamentous (and probably had a low percentage of fertility), as for instance after 50 days in *Pterygophora californica*, (McKay, 1933) or after 60 days in *Eisenia arborea* (Clare and Herbst, 1938).

Acknowledgements. This work is based on research supported by grants from the Deutsche Forschungsgemeinschaft to K.L., and to M.N. by the U.S. National Science Foundation (Grant No. OCE-76-24360) and the U.S. Department of Commerce (SGP 04-6-158-44110). Special thanks are due to Professor J.A. West and to Mr. W.N. Wheeler for helping collect the species from central California.

Literature Cited

Abbott, I.A. and G.J. Hollenberg: Marine algae of California, 827 pp. Stanford: Stanford University Press 1976

Anderson, E.K. and W.J. North: Light requirements of juvenile and microscopic stages of giant kelp, *Macrocystis*. *In:* Proceedings of the Sixth International Seaweed Symposium, pp 3-15. Ed. by R. Margalef. Madrid: Direccion de la Marina Mercante 1969

Branch, M.L.: Limiting factors for the gametophytes of three South African Laminariales. Investl Rep. Sea Fish. Brch S. Afr. *104*, 1-38 (1974)

Clare, T.S. and C.C. Herbst: The life history of *Eisenia arborea*. Am. J. Bot. *25*, 494-498 (1938)

Cole, K.: Gametophytic development and fertilization in *Macrocystis pyrifera*. Can. J. Bot. *46*, 777-782 (1968)

Cosson, J.: Action de la température et de la lumière sur le développement du gamétophyte de la *Laminaria digitata* (L.) Lam. (Phéophycée, Laminariale). C. r. hebd. Séanc. Acad. Sci., Paris (Sér. D) *276*, 973-975 (1973)

- Action des conditions d'éclairement sur la croissance des gamétophytes de *Laminaria digitata* (L.) Lamouroux (Phéophycée, Laminariale). Bull. Soc. phycol. Fr. *20*, 50-54 (1975)

- Action de la durée d'éclairement sur la morphogénèse des gamétophytes de *Laminaria digitata* (L.) Lam. (Phéophycée, Laminariale). Bull. Soc. phycol. Fr. *22*, 19-26 (1977)

-, P. Gayral and R. Jacques: Action de la composition spectrale de la lumière sur la croissance et la reproduction des gamétophytes de la *Laminaria digitata* (L.) Lam. (Phéophycée, Laminariale). C. r. hebd. Séanc. Acad. Sci., Paris (Sér. D) *283*, 1293-1296 (1976)

Druehl, L.D.: Taxonomy and distribution of northeast Pacific species of *Laminaria*. Can. J. Bot. *46*, 539-547 (1968)

Harries, R.: An investigation by cultural methods of the factors influencing the development of the gametophytes and the early stages of the sporophytes of *Laminaria digitata, L. saccharina,* and *L. cloustoni*. Ann. Bot. *46*, 893-928 (1932)

Hollenberg, G.J.: Culture studies of marine algae. I. *Eisenia arborea*. Am. J. Bot. *26*, 34-41 (1939)

Hsiao, S.I.C. and L.D. Druehl: Environmental control of gametogenesis in *Laminaria saccharina*. I. The effect of light and culture media. Can. J. Bot. *49*, 1503-1508 (1971)

- - Environmental control of gametogenesis in *Laminaria saccharina*. II. Correlation of nitrate and phosphate concentrations with gametogenesis and selected metabolites. Can. J. Bot. *51*, 829-839 (1973a)

- - Environmental control of gametogenesis in *Laminaria saccharina*. III. The effects of different iodine concentrations, and chloride and iodine ratios. Can. J. Bot. *51*, 989-997 (1973b)

- - Environmental control of gametogenesis in *Laminaria saccharina*. IV. *In situ* development of gametophytes and young sporophytes. J. Phycol. *9*, 160-164 (1973c)

Kain, J.M.: Aspects of the biology of *Laminaria hyperborea*. III. Survival and growth of gametophytes. J. mar. biol. Ass. U.K. *44*, 415-433 (1964)

- The biology of *Laminaria hyperborea*. V. Comparison with early stages of competitors. J. mar. biol. Ass. U.K. *49*, 455-473 (1969)

Kemp, L. and K. Cole: Chromosomal alternation of generations in *Nereocystis luetkeana* (Mertens) Postels and Ruprecht. Can. J. Bot. *39*, 1711-1724 (1961)

Larcher, W.: Physiological plant ecology, 252 pp. New York: Springer-Verlag 1975

Lüning, K. and M.J. Dring: Reproduction induced by blue light in female gametophytes of *Laminaria saccharina*. Planta *104*, 252-256 (1972)

- - Reproduction, growth and photosynthesis of gametophytes of *Laminaria saccharina* grown in blue and red light. Mar. Biol. *29*, 195-200 (1975)

Mathieson, A.C. and T.L. Norall: Physiological studies of subtidal red algae. J. exp. mar. Biol. Ecol. *20*, 237-247 (1975)

McKay, H.H.: The life-history of *Pterygophora californica* Ruprecht. Univ. Calif. Publs Bot. *17*, 111-148 (1933)

Mohr, H.: Lectures on photomorphogenesis, 237 pp. New York: Springer-Verlag 1972

Neushul, M.: The species of *Macrocystis* with particular reference to those of North and South America. Nova Hedwigia *32* (Suppl.), 211-222 (1971)

Pérez, R.: Influence de quelques facteurs physiques sur le développement de *Laminaria digitata* (L.) Lamour. Bull. Soc. phycol. Fr. *16*, 89-105 (1971)

Provasoli, L.: Media and prospects for the cultivation of marine algae. *In:* Cultures and collections of algae. Proceedings of the U.S.-Japan Conference, Hakone, September 1966, pp 63-75. Ed. by A. Watanabe and A. Hattori. Japanese Society of Plant Physiology 1968

Saito, Y.: An ecological study of *Undaria pinnatifida* Sur. I. On the influence of environmental factors upon the development of gametophytes. Bull. Jap. Soc. scient. Fish. *22*, 229-234 (1956a)

- An ecological study of *Undaria pinnatifida* Sur. II. On the influence of the environmental factors upon the maturity of gametophytes and early development of sporophytes. Bull. Jap. Soc. scient. Fish. *22*, 235-239 (1956b)

Sauvageau, C.: Sur le développement et la biologie d'une laminaire (*Saccorhiza bulbosa*). C. r. hebd. Séanc. Acad. Sci., Paris *160*, 445-448 (1915)

- Recherches sur les laminaires des côtes de France. Mém. Acad. Sci. Inst. Fr. *56*, 1-240 (1918)

Schreiber, E.: Untersuchungen über Parthenogenesis, Geschlechtsbestimmung und Bastardisierungsvermögen bei Laminarien. Planta *12*, 331-353 (1930)

Segi, T. and W. Kida: Studies on the development of *Undaria undarioides* (Yendo) Okamura. I. On the development of gametophytes and influence of light intensity on it. Rep. Fac. Fish. prefect. Univ. Mie *2*, 517-526 (1957)

Smith, H.: Phytochrome and photomorphogenesis, 223 pp. London: McGraw Hill Book Co. 1975

Sokal, R.R. and F.J. Rohlf: Biometry, 776 pp. San Francisco: Freeman 1969

Tseng, C.K., C.Y. Wu and K.Z. Ren: The influence of temperature on the growth and development of the Haidai (*Laminaria japonica*) gametophytes. Oceanologia Limnol. sin. *4*, 22-28 (1962)

Vadas, R.L.: Ecological implications of culture studies on *Nereocystis luetkeana*. J. Phycol. *8*, 196-203 (1972)

Yabu, H.: Early development of several species of Laminariales in Hokkaido. Mem. Fac. Fish. Hokkaido Univ. *12*, 1-72 (1964)

Dr. K. Lüning
Biologische Anstalt Helgoland
(Meeresstation)
D-2192 Helgoland
Germany (FRG)

Date of final manuscript acceptance: September 23, 1977. Communicated by O. Kinne, Hamburg