# Ecological Differences Between the Isomorphic Phases of *Mazzaella lilacina* (Rhodophyta, Gigartinaceae): 1) Spore Production, 2) Recruitment Specialization, 3) Resistance to Removal By Wave Action.

By

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## Abstract

Some of the potential factors responsible for the observed increase in the proportion of tetrasporophytes in wave exposed populations of *Mazzaella lilacina* (Postels et Ruprecht) Leister have been examined.

By counting the number of spores per reproductive structure tetrasporophytes were found to hold significantly more spores than gametophytes in August, 1990. Only slightly more tetraspores than carposspores were found at other sampling time ($p=0.127$ and 0.637).

As expected, based on the production of spores, the proportion of gametophytes among recruits into intertidal clearings was high , although not as high in the exposed site as was expected based on the proportion of reproductive tetrasporophytes in the population at the time the clearings were made suggesting that tetrasporophyte recruitment is somehow favoured. The opposite was observed in the sheltered site suggesting that gametophyte recruitment is favoured in that site.

Monitoring of blades following recruitment showed a slight trend towards a decreasing proportion of gametophytes in the exposed site and an increasing proportion of gametophytes in the sheltered site, suggesting that the mortality of on phase was higher than the other in each site.

Finally, thalli of *M. lilacina* were pulled and broken in the field . Force to break and surface area data was used to generate curves showing the proportion of each phase which would be broken with water moving at velocities from 0 to 20 m/s. Gametophyte blades longer than 15cm in the exposed site were significantly more susceptible to removal by moving water than were tetrasporophyte blades.

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## Introduction

This thesis explores the mechanisms responsible for changes, with wave energy, in the proportions of the phases of Mazzaella lilacina (Postels et Ruprecht) Leister, as noted by Dyck et al. (1985). Others (Mathieson and Burns 1975) have reported similar trends for Chondrus crispus Stackhouse in New Hampshire, U.S.A..

M. lilacina has a triphasic sporic life history with isomorphic haploid male and female gametophytes and a diploid tetrasporophyte. Several annual blades develop from each perennial holdfast. Following fertilization of the egg the diploid carposporophyte generation develops in the female gametophyte blade. The carposporophyte consists of carposporangia and carpospores and is surrounded by the cystocarp, made up of haploid female tissue. The whole structure forms a small bump on the surface of the female gametophyte blade.

Following their release and germination carpospores develop into tetrasporophytes. At the onset of reproductive maturity many small bumps, sori, form on the surface of the tetrasporophyte blade. The bumps formed by cystocarps are generally larger and less densely packed than the tetrasporangial sori. Throughout this thesis 'bumps' will be used to refer to reproductive structures in general. The size and density of bumps form the primary differentiating characteristics between the female gametophytes and tetrasporophytes in the field. Male gametophytes remain smooth throughout their lives. A slight colour change indicating the onset of reproduction is the only means of field identification.

Littler and Littler (1980) proposed a model in which algae having similar morphologies will also behave in a similar manner ecologically. A comparison of blade thickness and photosynthetic rate for a number of taxa, supported their premise. If Littler and Littler's 'Functional/Form' model was correct then in populations of isomorphic algae the proportions of the phases should approach a 1:1 ratio (May 1986). If one phase became more abundant a greater proportion of the spores released into the water column would be from the numerically dominant phase, leading to an increase in the less abundant phase. However, it has been well documented for the isomorphic phases of a number of algal taxa, including Mazzaella, that ecological differences between the phases do exist, so the 'Functional/Form' model clearly is not appropriate in all situations.

Luxoro et al. (1989) and Buschmann (1991) both reported for Mazzaella laminarioides (Bory) Fredericq (as Iridaea Laminarioides) that some herbivores had a feeding preference for one phase over the other. Destombe et al. (1992) found that haploid spores of Gracilaria verrucosa (Hudson) Papenfuss floated longer and had better survival under different light conditions. It was also reported (Destombe et al. 1993) that holdfasts of G. verrucosa from haploid juveniles grew better than those from diploid juveniles under non-optimal conditions. For Chondrus crispus Bhattacharya (1985) measured faster growth rates for gametophytes compared to tetrasporophytes during most of the study period.

Although the mechanism is unclear, one well documented difference between the phases of Mazzaella lilacina is in their timing of peak blade density and peak reproductive blade density (DeWreede and Green 1990, Dyck et al. 1985, Hansen and Doyle 1976, Hansen 1977, Hannach and Santelices 1985 for Mazzaella laminarioides). The phases of M. lilacina have been shown to follow a regular annual alternation of numerical dominance in some populations (DeWreede and Green 1990, Dyck et al. 1985). Gametophytes have their peak in abundance in the spring and early summer followed by tetrasporophyte numerical dominance in the fall and winter. Senescence of the gametophytes occurs earlier in the year than senescence of the tetraporophytes. By mid-winter virtually all of the gametophyte blades are gone, whereas a few tetrasporophyte blades live until spring. While, Hansen and Doyle (1976) and Hansen (1977) did not find an alternation of numerical dominance they did find that the peak in reproductive gametophyte abundance and biomass, though lower at all times than the peak for reproductive tetrasporophytes, was in the spring-summer; while the peak in reproductive tetrasporophyte abundance and biomass was in the fall. The temporal separation in peak blade densities indicates that some important underlying differences exist between the free living phases of Mazzaella lilacina.

A practical aspect of phase cycling is that one sampling time is not sufficient to characterize a population. Unless the questions asked are time specific, comparisons between the phases must be made over the course of at least one year or preferably several, with several sampling times within each year.

One explanation for the offset peaks in phase abundance is based on different growth rates for gametophytes and tetrasporophytes and has been tested in a computer model simulating a population of Mazzaella lilacina in Vancouver Harbour, B.C.(Ang et al. 1990). If gametophytes grow faster then even if juveniles of both phases initiate growth at the same time the gametophytes will reach their reproductive and density peak sooner than the tetrasporophytes.

As well as an annual cycling in abundance peaks for the gametophytes and tetrasporophytes it has been reported (Hannach and Santelices 1985) that the proportion of tetrasporophytes in populations of Mazzaella laminarioides (as Iridaea laminarioides), in Chile, increases with depth. A similar pattern has been reported for Chondrus crispus (Mathieson and Burns 1975, Craigie and Pringle 1978 ); the proportion of tetrasporophytes was reported to increase with depth and distance from shore. It should be noted that Mathieson and Burns (1975) used only one sampling time and did not analyze their data statistically, thus the sampling error is not known. Lazo et al. (1989) reported the same distributional pattern as Craigie and Pringle (1978) but only within the same limited range of depths; in a broader sample no pattern was apparent. So far no hypotheses have been tested which explain the observed distributions of the phases in relation to depth or wave exposure.

Work has been done comparing growth optima and growth rates of adult gametophytes and tetrasporophytes. Hansen (1977) reported similar growth rates for both phases of a Mazzaella lilacina (as I. cordata) population, in California. Hansen (1980) also reported that the photosynthetic light saturation levels for both phases was approximately 150 micro E $m^2$ $s^{-1}$. May (1986) on the other hand found that the summer growth rate (June), expressed as change in blade area per day, for tetrasporophytes was significantly higher than for gametophytes in a *Mazzaella lilacina* (as *Iridaea cordata*) population on San Juan Island, Washington. However, the results do not necessarily mean that tetrasporophytes generally grow faster than gametophytes because growth rate was measured so late in the spring. The slower growth rate of gametophytic plants may be attributable to an already mature gametophyte population which had already slowed its growth (May 1986).

Another difference between isomorphic gametophytes and tetrasporophytes, which may be reflected ecologically, has been demonstrated for several species of the Gigartinales. Carrageenans are sulphated polysaccharides, which comprise much of the cell walls of both gametophytes and tetrasporophytes (McCandless et al. 1975, 1983). Gametophytes have gelling Kappa carrageenan and tetrasporophytes contain viscous, non-gelling Lambda carrageenan. The chemical difference between the carrageenans is the basis for the resorcinol-acetal test, which is used for differentiating between vegetative thalli of the two phases. The Kappa carrageenan found in gametophytes contains 3,6-anhydrogalactose which turns the resorcinol-acetal reagent red, whereas, Lambda carrageenan lacks this component and causes no colour reaction (Shaughnessy and DeWreede 1991).

Clearly there are ecological differences between the isomorphic phases of *Mazzaella lilacina*. The mechanisms responsible for the separations between the gametophytes and tetrasporophytes in time and space have yet to be tested. Three possible mechanisms responsible for the observed increase in the proportion of tetrasporophytes with wave exposure will be examined in the following chapters: Chapter 2) Spore production; do cystocarpic and tetrasporic blades produce similar numbers of spores? Chapter 3) Recruitment; within one wave exposure regime, do the two isomorphic phases recruit in similar proportions? And, are the phase proportions of recruits different in areas subject to different amounts of wave energy? Chapter 4) Biomechanics; are tetrasporophytes more resistant to removal by drag forces generated by moving water?

If either the gametophytes or tetrasporophytes produce more spores than the other then one would expect recruitment to reflect the bias in spore availability. A shift away from the expected recruitment proportions would indicate that some source of phase discriminate mortality was active between spore production and recruitment. Following recruitment, if the proportions of gametophytes and terasporophytes changes as the thalli age then some source of phase discriminate mortality must be at work. Sources of blade mortality will not be examined in this research, except the mortality due to water motion.

Field work was conducted at two sites on opposite sides of a 25m long peninsula in Barkley Sound, British Columbia. These sites were chosen because of the obvious differences in wave exposure to which they were subjected. One site, referred to as 'exposed', faces directly into waves approaching Second Beach: the other site referred to as 'sheltered', faces the beach and therefore is protected from the direct action of waves. The exposed site is near the wave exposure extreme for Mazzaella lilacina (Shaughnessy Pers. Comm.). Other floristic differences between the two sites are described in Chapter 1, including comparisons of the taxa found in each site and elevation comparisons of taxa found in both sites.

# Chapter 1: Community Analysis

## Introduction

All of the research in the following chapters is based on population sampling from two areas in Barkley Sound, British Columbia which are subject to very different amounts of wave energy. The purpose of this chapter is to describe some of the biological differences between the two study areas. Some organisms were found only in one site or the other and some of the common organisms were present at different intertidal heights in each area. Information came from two sources; the first was a survey of the flora and fauna from both sites in the summer of 1992, the second was recruitment data from clearings which were made at different times of year and monitored for up to two years. Chapter 3 is a comparison the recruitment of the gametophytes and tetrasporophytes of *Mazzaella lilacina* into the sheltered and exposed sites. The clearings mentioned above were made for the *Mazzaella lilacina* recruitment study, but were used as well to monitor the recruitment of other algae and invertebrates.

Because of the sporadic occurrence of many of the organisms statistical methods were not used, instead, the information was presented using bar graphs.

## Materials and Methods

A grid, 6 m by 10 m, made of thin string was placed in each of the study sites and held in place by concrete nails hammered into the rock. Randomly selected coordinates were used to place each 0.25 m square quadrat within the grids. Within the single sampling event, a total of 65 quadrats were sampled, 33 in the sheltered site and 32 in the exposed site. The percent cover for every species of algae and group of invertebrates was estimated. Multilayered quadrats could have percent covers of more than 100%.

The heights of 12 points on each grid were measured, in relation to the tide height at the time, using a simple water level. Using the time at which the measurement were taken the tide height could be found and the height measurements changed to use Canadian Chart Datum as their reference. The height of each sampled quadrat was interpolated from the measurements on each grid taken in the field. A water level consists of a meter stick, a bucket containing a known depth of water, and a length of clear plastic hose. The meter stick was placed vertically at the waters edge with one end of the plastic hose running up the length of the stick. The other end of the plastic hose was placed in the bucket full of water. By sucking on the end of the hose attached to the meter stick water was drawn out of the bucket into the hose. Gravity moved the water through the hose and up the length of the meter stick until it reached the same elevation as the top of the water in the bucket. The elevation of any point could be determined by placing the bucket on the point, water would flow into or out of the hose until the top of the water in the bucket and the water in the meter stick end of the hose were at the same elevation, at which point the elevation above the tide level could be read off of the meter stick. The immediate reference point for this procedure was the tide height at the time the measurements were taken. Using the Canadian tide tables the height above Canadian Chart Datum could be determined from the time at which the measurements were taken.

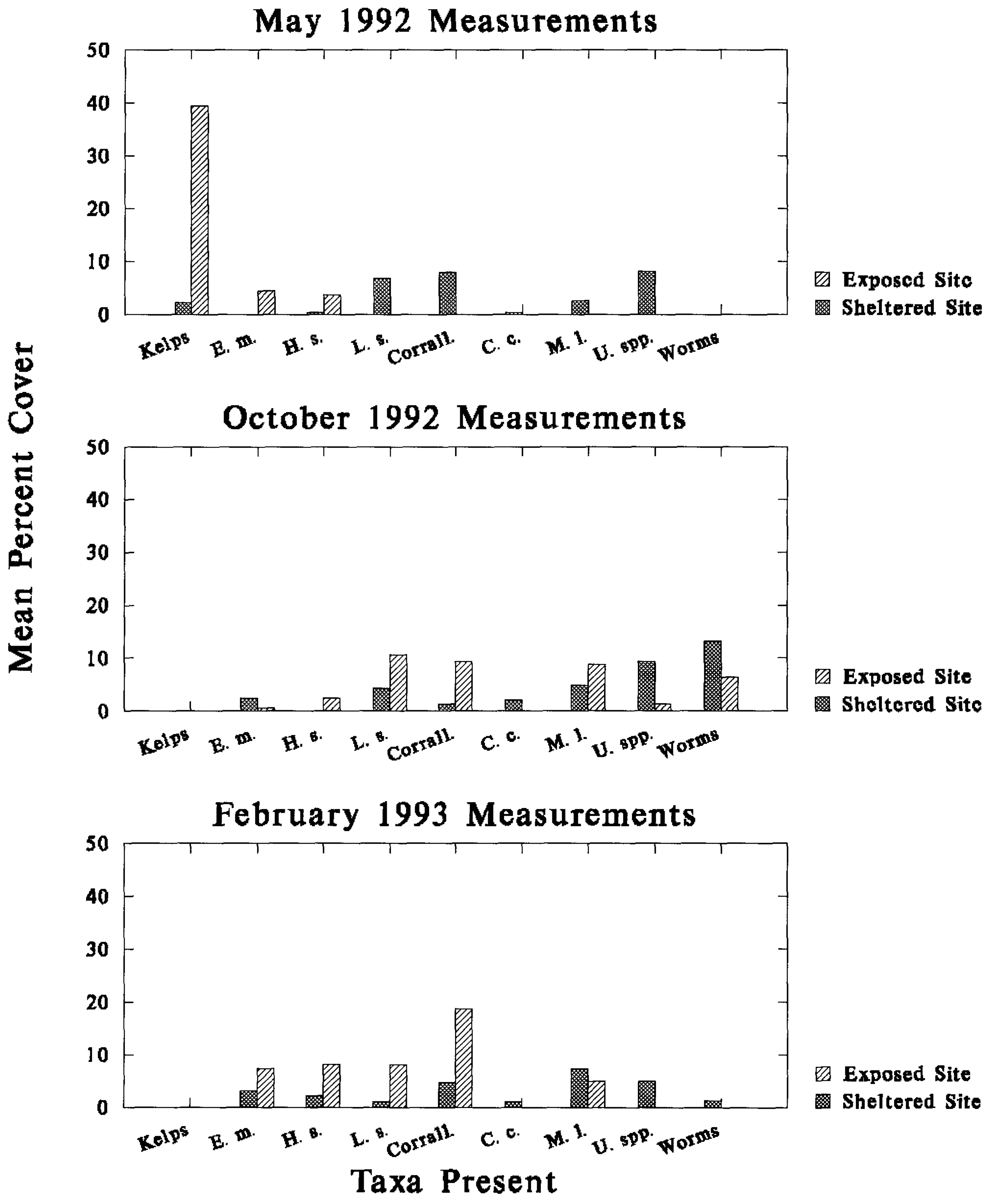
As part of the recruitment work in chapter 3, several clearings were made within both sites at three different times of year and the recruitment followed for several months. For a complete explanation of the sampling method see the Materials and Methods section of chapter 3. Following creation of the clearings percent cover of each type of algal colonizer was monitored approximately every two months until the Fall of 1993. Taxa which were rarely found were not included in the comparisons of the sheltered and exposed areas. A height measurement was taken from each clearing using the water-level method discussed in the section above.

Due to the sporadic occurrence of most of the taxa, statistical procedures were not used, instead, the data have been presented using bar graphs.

## Results

Colonization of experimentally cleared plots in the two sites, shown as mean percent cover, is presented in Figures 1.1-1.3. Colonizers were grouped as follows: 1) Kelps, which refers to juvenile of several genera which were difficult to distinguish from one another. As the thalli matured they were placed into more specific groups and the general Kelps group disappeared. 2) Egregia menziesii (Turner) Areschoug., 3)Hedophyllum sessile (C.A. Agardh) Saunders, 4) Laminaria setchellii Silva, 5) Crustose corrallines, 6) Chondrochanthus corymbiferus (Kutzing) Guiry, 7) Mazzaella lilacina (Postels and Ruprecht) Leister, 8) Ulva spp., and 9) tiny calcareous tube worms.

Figure 1.1: Colonization of clearings made in November 1991. Measurements taken in May 1992, October 1992, February 1993, and October 1993 are shown.



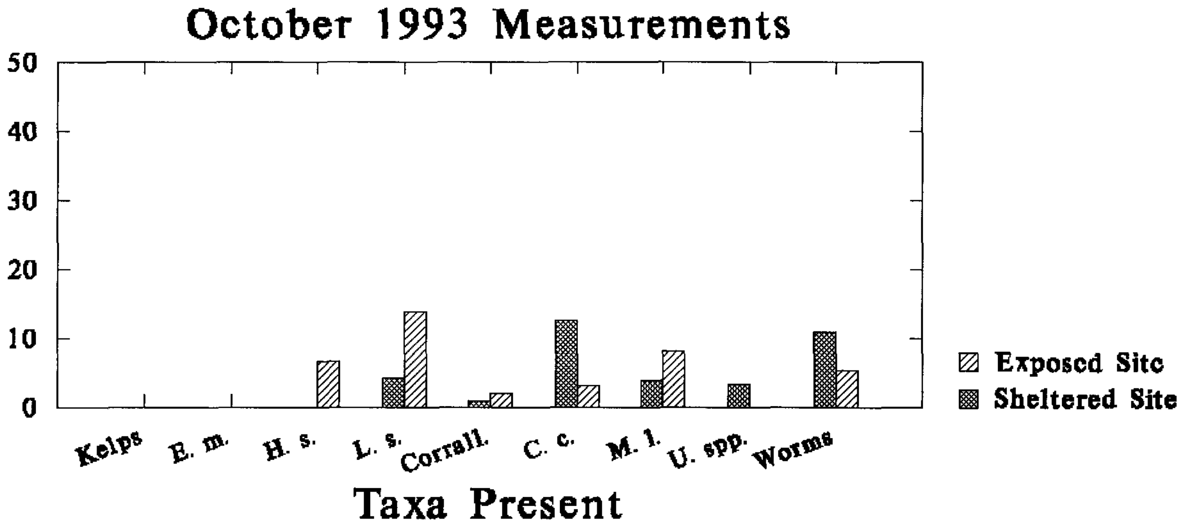

October 1993 Measurements
50
40
30
20
10
0
Kelps
E. m.
H. s.
L. s.
Corrall.
C. c.
M. l.
U. spp.
Worms
Taxa Present
Exposed Site
Sheltered Site

Figure 1.2: Colonization of clearings made in May 1992. Measurements taken in October 1992, February 1993 for the Exposed site only, April 1993 for the Sheltered site only, and October 1993.

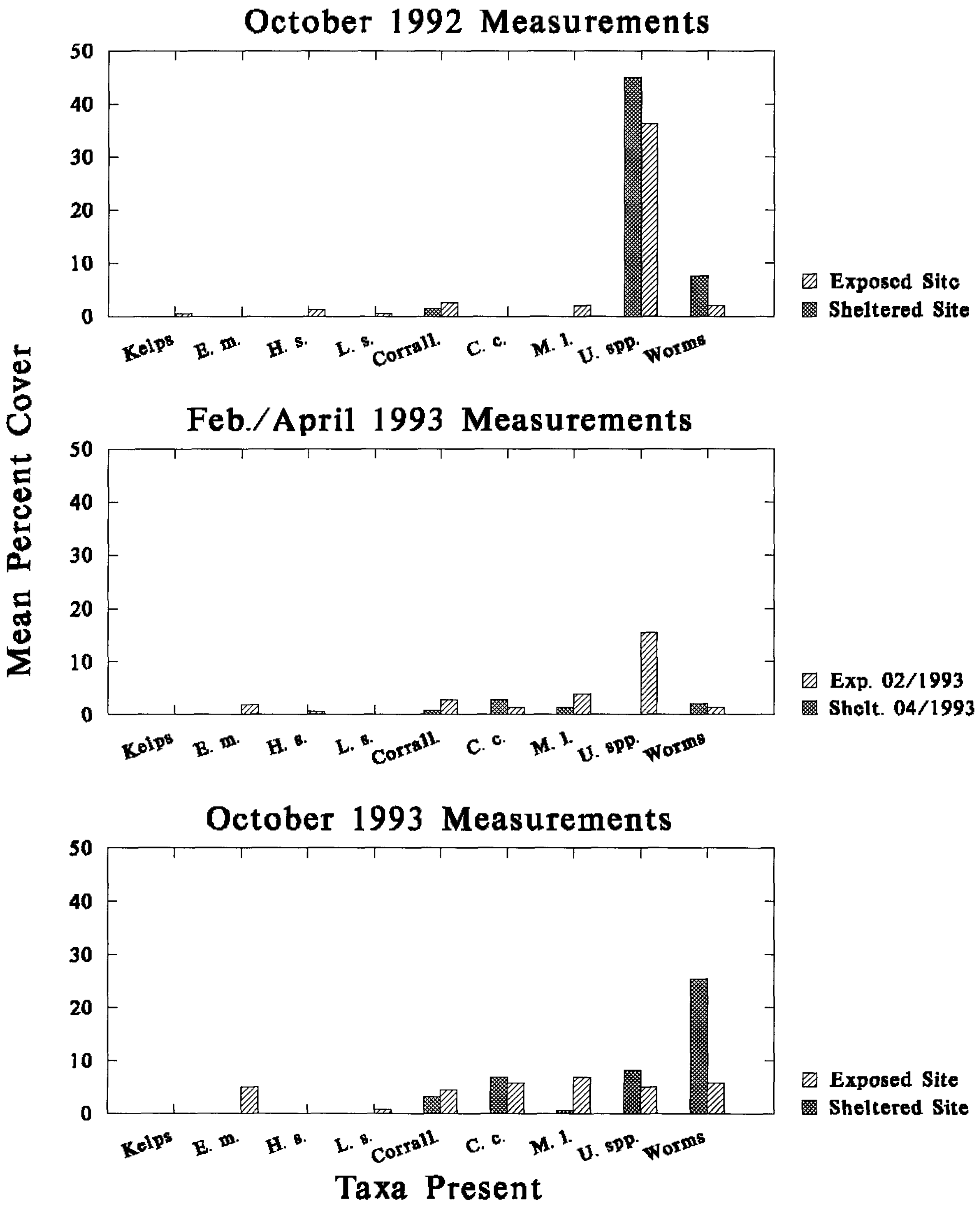
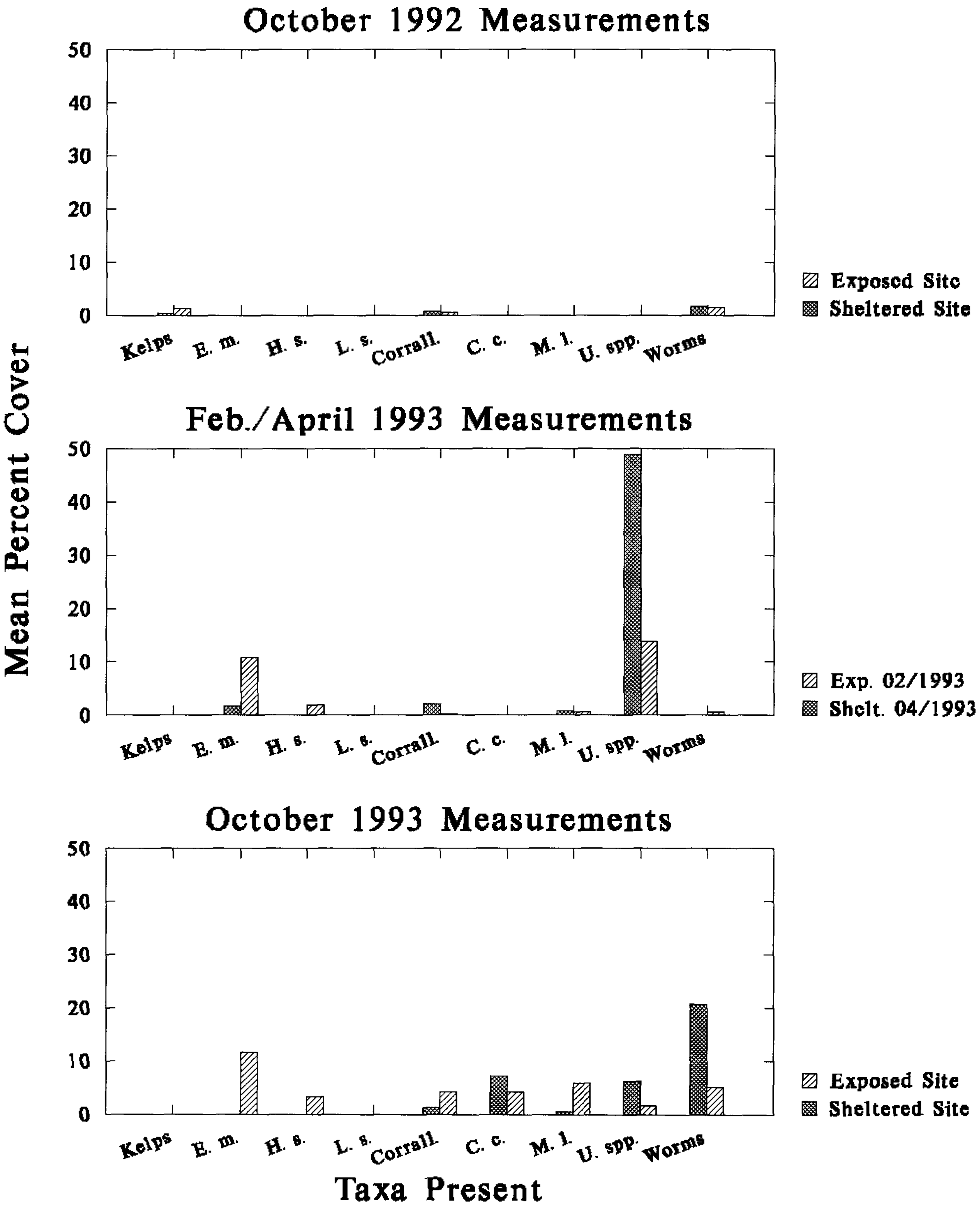


Figure 1.3: Colonization of clearings made in July 1992. Measurements taken in October 1992, February 1993 for the Exposed site only, April 1993 for the Sheltered site only, and October 1993.



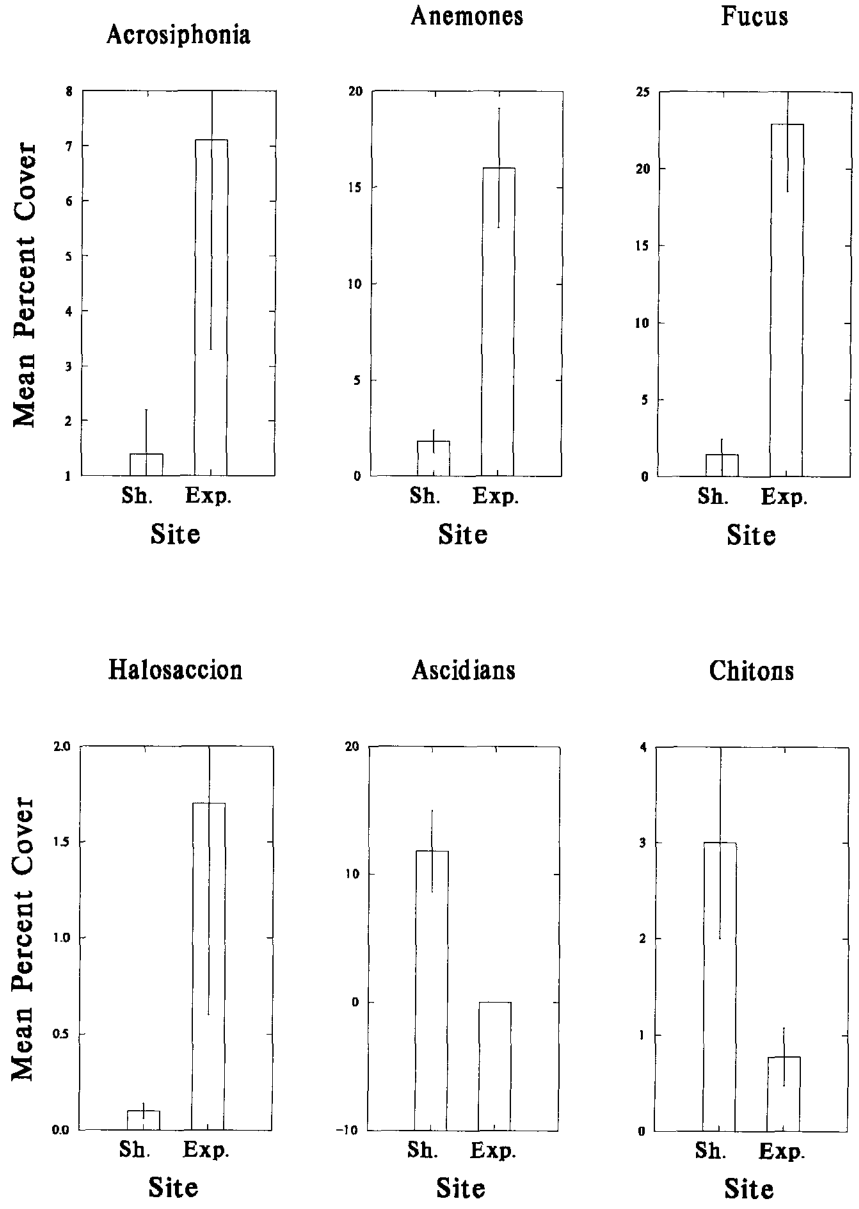
In the November 1991 clearings in the May 1992 sampling time (Figure 1.1), while both the sheltered and exposed sites experienced some recruitment, the taxa recruited were not shared between the two areas. Five months later, in October 1992, most taxa were common to both sites, as was also true for the last two measurement dates. Chondrocanthus corymbiferus was restricted to the sheltered site until sometime after February 1993. Ulva was also only present in the sheltered area, except for a small amount in October of 1992. It should also be noted that while Mazzaella lilacina recruited first into the sheltered area later measurements showed the percent covers in the two sites to be similar.

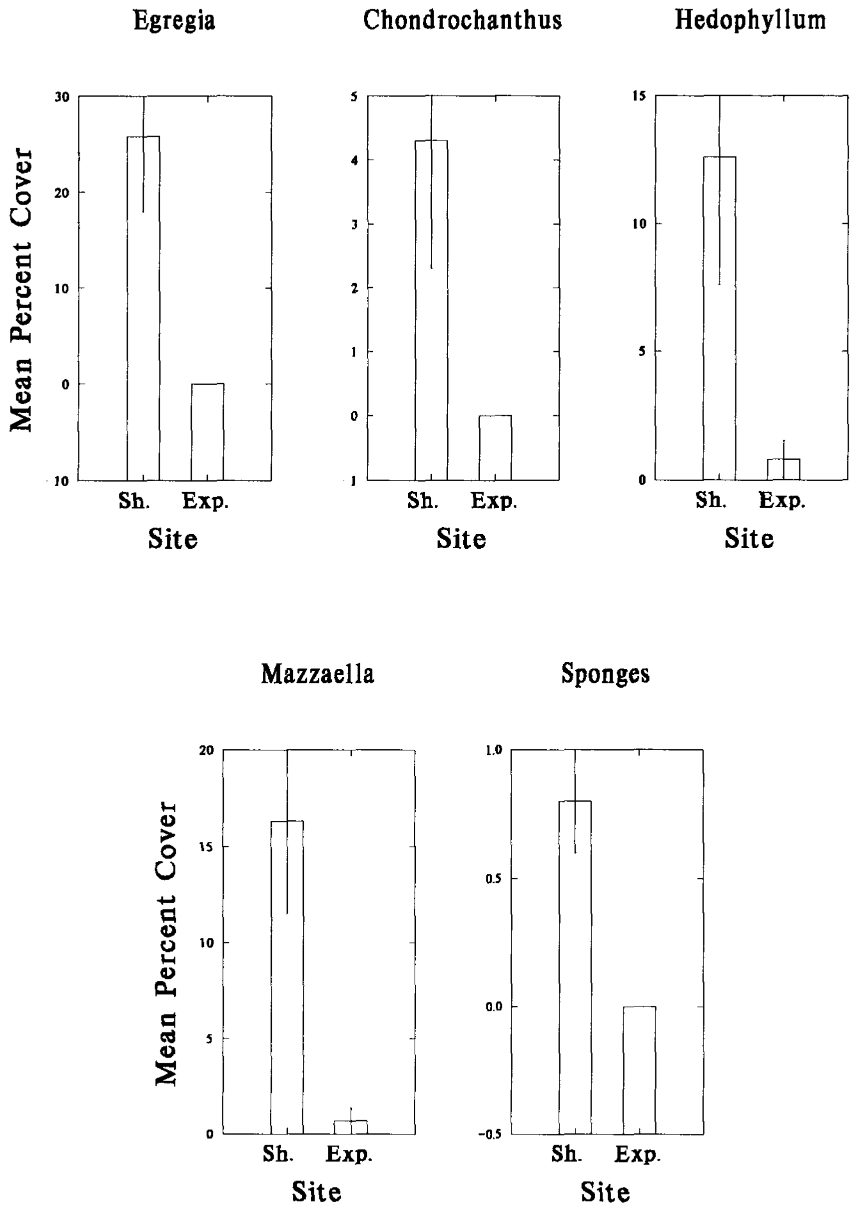
Two trends were immediately apparent for the colonizers of the May 1992 clearings (Figure 1.2), especially when compared with the earlier clearings. The first was the huge abundance of Ulva spp. in both sites. The second was the nearly complete absence of kelps. Corresponding with the decrease in abundance of Ulva spp.was an increase in the abundance of other taxa. Most taxa were present in similar amounts in both sites by the Spring of 1993, with the exception of kelps which didn't recruit into the sheltered clearings at all. The abundances of Mazzaella lilacina were also notably different in the two sites in the October measurement time, with the thalli in the exposed site covering much more area (i.e. larger or more abundant).

In the July 1992 clearings (Figure 1.3) the most striking recruitment was by Ulva spp.. Growth of other taxa followed the same pattern as in Figure 1.2: as the abundance of Ulva spp. decreased the abundance of other taxa increased. The two processes were not necessarily causally related. However, again there was nearly a complete absence of kelps from the sheltered area. Laminaria setchellii was absent from both sites at all measurement dates. As well, *Mazzaella lilacina* covered much more area in the exposed clearings than in the sheltered clearings.

The most obvious difference between the sites was in the abundance of kelp recruits. Generally the kelps covered much more area in the exposed clearings than in the sheltered clearings. *Ulva* spp., however, generally covered much more area in the sheltered site clearings. *Mazzaella lilacina* covered similar areas in the November 1991 clearings in both sites, but covered much more area in the exposed site than the sheltered site in the spring/summer clearings.

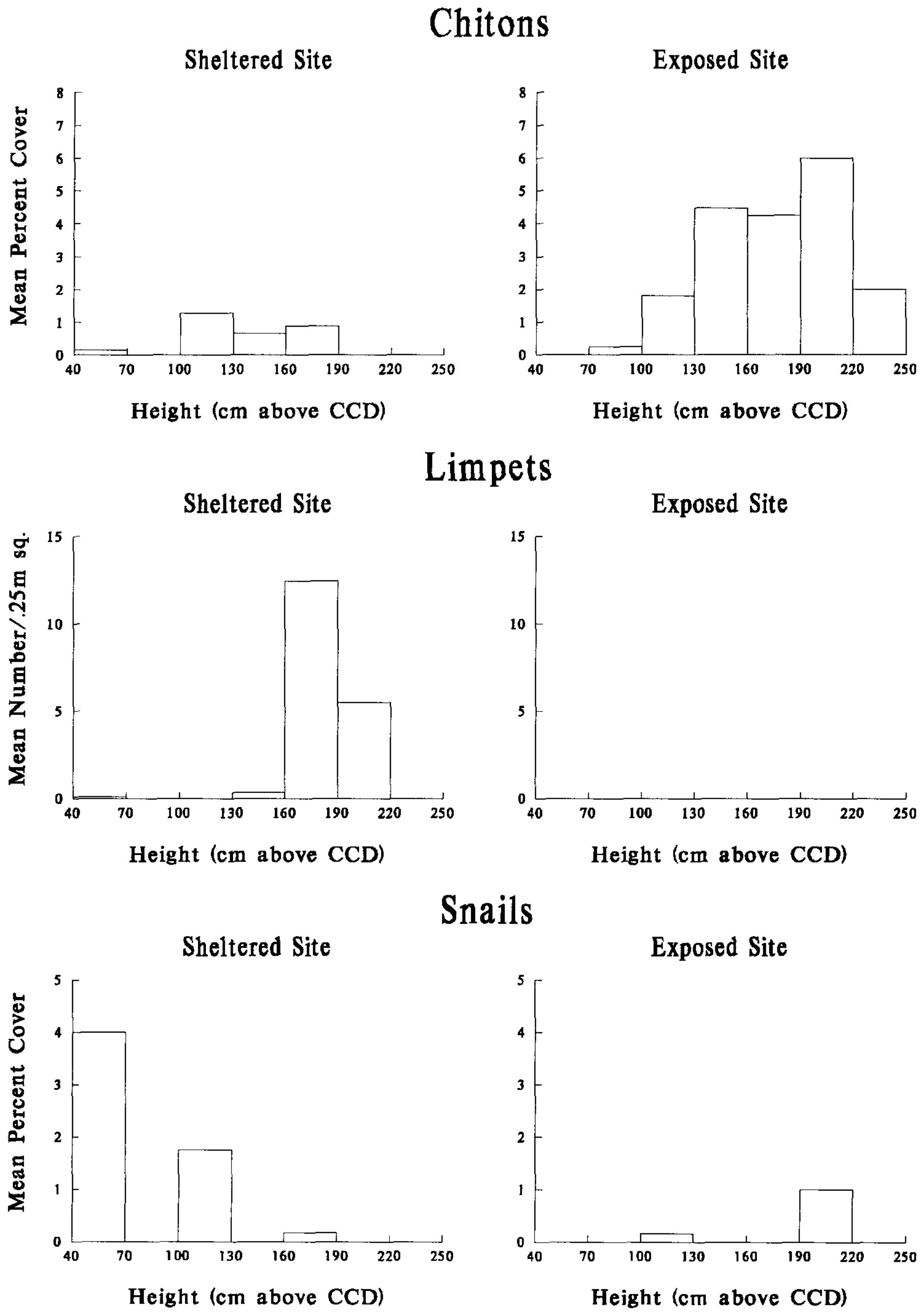
Figure 1.4: Data from the community survey. The taxa in the first four graphs were much more abundant in the sheltered area than in the exposed area, whereas the opposite was true for the following seven. Each bar is surrounded by error bars indicating standard error. Elevations within the intertidal zone of 80 to 220 cm above Canadian Chart Datum were used for both areas. Of the 60 taxa found in the survey 11 were selected which were found frequently within at least one of the study areas.

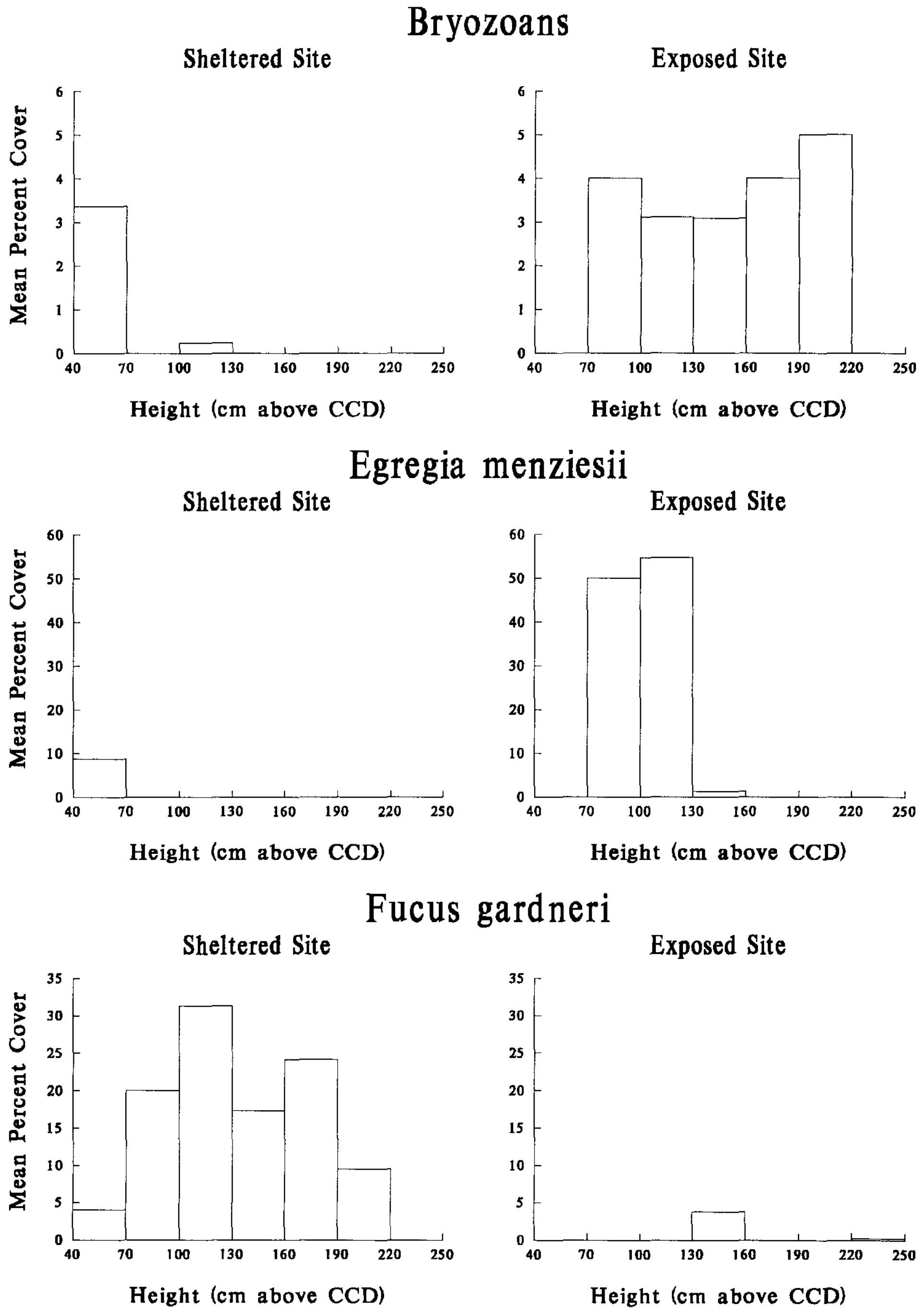



Egregia
Chondrochanthus
Hedophyllum
Mean Percent Cover
30
20
10
0
10
5
4
3
2
1
0
1
15
10
5
0
Sh.
Exp.
Site
Mazzaella
Sponges
20
15
10
5
0
1.0
0.5
0.0
-0.5

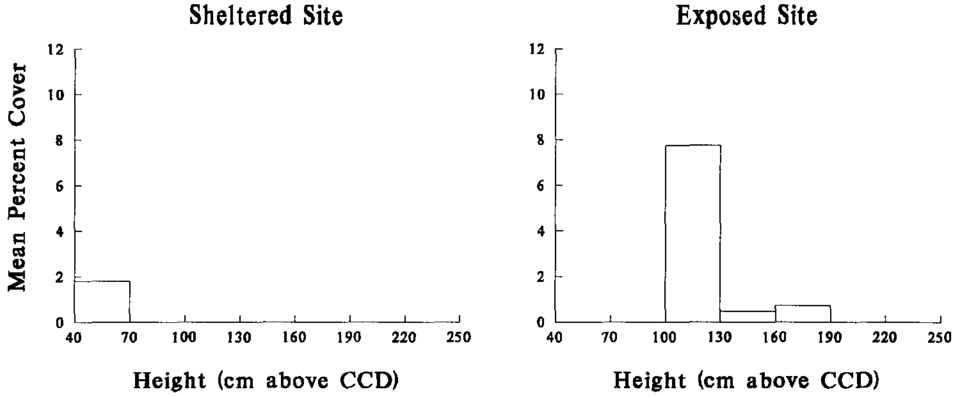
*Acrosiphonia* spp., aggregating anemones, *Fucus gardneri* Silva, and *Halosaccion glandiforme* (Gmelin) Ruprecht were all found more frequently in the sheltered site. The opposite was true for Chitons, *Hedophyllum sessile*, and *Mazzaella lilacina*. Ascidians, *Chondrocanthus corymbiferus*, *Egregia menziesii*, and sponges were completely absent from the sheltered site but were frequently found in the exposed area. In some cases taxa which appeared to be completely absent from an area may have just been absent within the elevation range used for this comparison.

Figure 1.5: A comparison of the elevation distributions of a number of taxa. Within the sheltered area measurements were taken between 40 and 220 cm above Canadian Chart Datum; for the exposed area the range was between 80 and 250 cm above CCD.

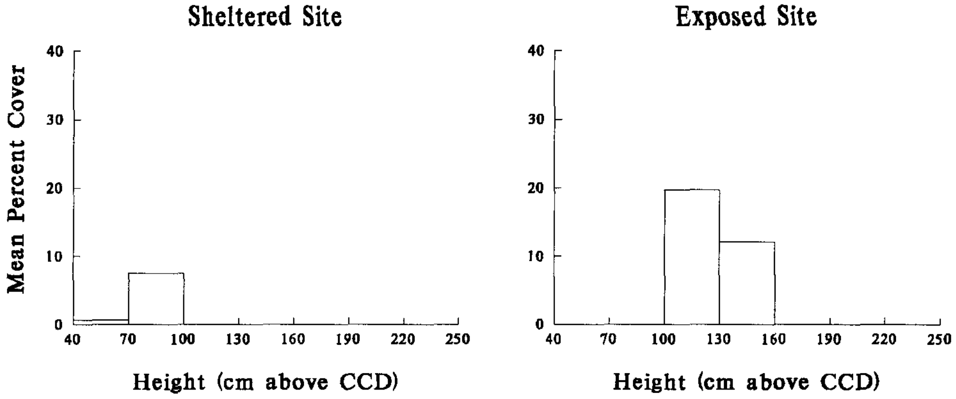



Bryozoans
Sheltered Site
Exposed Site
Mean Percent Cover
0
1
2
3
4
5
6
40
70
100
130
160
190
220
250
Height (cm above CCD)
Egregia menziesii
Sheltered Site
Exposed Site
Mean Percent Cover
0
10
20
30
40
50
60
Height (cm above CCD)
Fucus gardneri
Sheltered Site
Exposed Site
Mean Percent Cover
0
5
10
15
20
25
30
35
Height (cm above CCD)

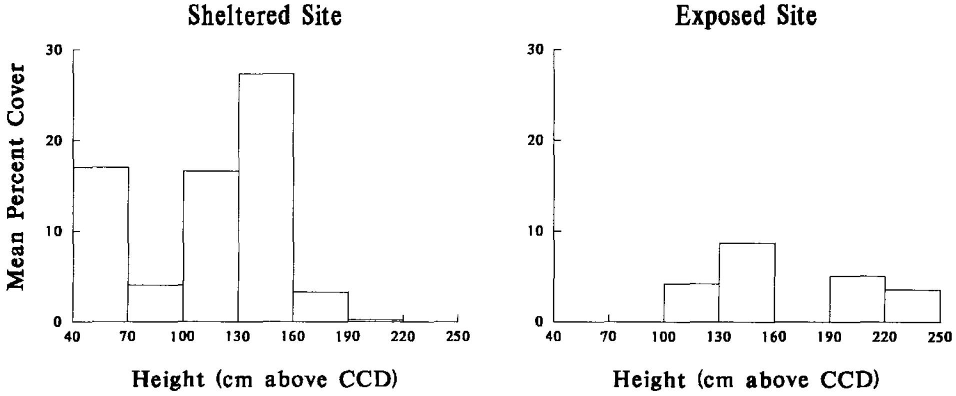
Chondrocanthus corymbiferus



Mazzaella lilacina



Ulva spp.



No general trend in herbivore abundance or elevation preference was apparent. Within both areas chitons had a similar elevation range, but extending into the higher elevations in the more wave exposed area. The elevation range of chitons in the sheltered site did not overlap with the elevation range for M. lilacina. In the wave exposed area, however, the top of the M. lilacina zone overlapped with the bottom of the chiton zone. Limpets were completely absent from the wave exposed survey area and were restricted to the top of the study elevation range in the sheltered area. Snails, mostly Littorina spp., covered the greatest area near the bottom of the intertidal zone in the sheltered site and strongly overlapped with M. lilacina. In the wave exposed area, however, very few snails were present in the same elevation range as M. lilacina.

While Fucus and Ulva were found in both sites they both had a wider range in the more sheltered area, extending further down the shore. For all other algal taxa and bryozoans the bottom of the study range was occupied in both study sites but they extended further up the shore in the more wave exposed area.

## Discussion

Figures 1.1-1.3 demonstrate how variable the recruitment of species following clearing can be. In the winter clearing *Egregia* recruited into both sites, whereas in the spring and summer clearings it recruited only into the exposed area. *Ulva*, on the other hand, recruited equally well into both areas in the spring and summer clearings but was nearly excluded from recruiting into the Winter clearings in the wave exposed area. Recruitment of *Mazzaella lilacina* followed a pattern similar to that for *Egregia*, in the Winter clearings recruitment was similar in the two areas, but in the spring and summer clearings the amounts were much greater in the exposed area. Recruitment success is a complex mix of propagule availability, propagule site selection, biological interactions such as competition and predation, and physical factors including dessication, irradiance, temperature, salinity, and wave energy (Underwood and Denley 1984). To attribute observed recruitment to any simple set of interactions without further study would not further our understanding of the system

The virtual absence of the first four taxa in Figure 4 from the more wave exposed site may be only an indirect result of higher wave energy. For example, *Acrosiphonia* grows as a small tuft in cracks and is not likely to be subjected to much direct force from moving water. More likely explanations for its absence involves herbivores, which also tend to live in cracks, and possibly the availability of propagules for recruitment. *Halosaccion*, on the other hand, has a morphology which is likely directly influenced by water motion. *Halosaccion* grows as a water filled, tubular sac with a small holdfast. Taxa 5-9 were all larger or more abundant in the wave exposed area. Food availability may be an important factor for the sessile invertebrates like sponges and ascidians. The water in the more sheltered area may not supply enough food. Again, without more study it is impossible to attribute causal relationships between biotic or abiotic factors and the presence or absence of certain taxa. It was apparent, however, that Mazzaella lilacina was much more successful at growing in the more wave exposed environment than it was in the sheltered area. The area covered was many times higher in the exposed area, at a time of year when blade density is high. The site comparisons in this figure do not take into consideration phase proportions.

One general trend was apparent in Figure 1.5. In almost all cases where a taxon was shared between the sheltered and exposed areas its range was higher in the intertidal zone in the more wave exposed area (Underwood 1981, Chapman 1986). Although other factors may be as or more important, the most obvious explanation for the elevation shift in the two sites is decreased desiccation and temperature in the site subject to more wave action. Exceptions to the above trend were snails, Fucus gardneri, and possibly Ulva spp.. In these cases the distributions had similar upper limits, but extended lower in the sheltered area.

If herbivores are important in the creation of different phase proportions of Mazzaella lilacina in the two areas then chitons and snails are likely candidates, and deserve further study. While many limpets are present in the sheltered area they were not found in the Mazzaella lilacina zone. A preference by both or one herbivore for one phase over the other could help to account for the increased proportion of tetrasporophytes in the more wave exposed site.

# Chapter 2: Spore Production

## Introduction

As discussed in the Introduction, there is accumulating evidence that there are ecological differences between the isomorphic phases of various red algae. In Mazzaella lilacina the proportion of tetrasporophytes in populations tends to increase as wave energy increases. No mechanisms have been tested which account for this. This chapter examines the numbers of spores produced by gametophytes and tetrasporophytes of Mazzaella lilacina in a wave exposed population. Two questions will be asked: 1) do gametophytes and tetrasporophytes produce similar numbers of spores per blade? 2) is there a peak in the number of spores per reproductive structure within each phase?

A regular cycling of reproductive blade densities has been well documented (Hansen 1976, DeWreede and Green 1990, Dyck 1991) for the isomorphic phases of Mazzaella lilacina. In the study site cystocarpic blades are present during most of the year, with their lowest abundance occurring in the winter (Shaughnessy Pers. Comm.). In contrast reproductive tetrasporophytes occur in significant numbers throughout the year. The peak of reproductive blade density occurs in the late spring and summer for cystocarpic blades and in the fall for the tetrasporophytes. Information on the production and release of spores is much more sketchy. Estimates of spore release have been made for several species of algae including Mazzaella lilacina (as I. cordata), Chondrus crispus and Gelidium robustum (Green 1989, Bhattacharya 1985, and Melo and Neushul 1993). In all cases the thalli were induced to release their spores under culture conditions over a few days. Rapid degeneration of the thalli in some cases limited the length of the release period. It is not known how accurately spore release studies have reflected natural populations. Counting the spores present within mature sori or cystocarps gives a more accurate estimate of spore production, but it is not possible to be certain that all of the spores produced are released. Many sori and cystocarps do not release their spores before the blades senesce (Pers. Obs., Luxoro et al. 1989).

No comparisons have been made between cystocarpic and tetrasporic blades with respect to the number of spores produced or released at different times of year. One comparison (Green, MSc Thesis 1989) looked at the numbers of spores released per unit of blade surface area in September. She reported that many times more carpospores were released than tetraspores. As well, no comparisons have been made between the numbers of carpospores or tetraspores released in different months. Melo and Neushul (1993) compared the number of tetraspores released by Gelidium robustum at several times of the year. They found that although the most sori per gram of tissue was found in the spring and summer the highest release rate per sorus per day was found in the winter. They also found that spore germination was about six times more successful in the spring/summer than in the winter. Green (1989) reported the opposite trend in sori density for a population of Mazzaella lilacina (as Iridaea cordata) in Vancouver Harbour. A greater density of sori were found in January than in August. Comparisons of spore release rate were not made.

It is not known for Mazzaella lilacina whether the number of spores produced by one phase varies significantly over the course of a year, and if so when the peaks occur. Does the peak in spore production correspond with the peak in reproductive blade density? As well, it is unknown whether there is a period during the annual cycling of reproductive blade densities when one or both spore producing phases of M. lilacina produce more spores than the other. The timing of spore production peaks could help to explain the observed spatial phase distributions, tetrasporophyte dominance being explained by the timing of clearing events in relation to the peak in carpospore production.

The questions to be addressed in this chapter are: 1) do gametophytes and tetrasporophytes produce comparable numbers of spores per blade? 2) is there an annual pattern in the number of spores per reproductive structure within each phase?

## Materials and Methods

The thalli used in this study were selected from the wave exposed site, using an haphazardly placed transect with randomly spaced marks on it. The transect was placed within the intertidal M. lilacina zone and one blade from each holdfast within a quarter meter square quadrat was collected. Approximately 35 quadrats were sampled in total. Collections were made as part of population studies in 1990/91 by Frank Shaughnessy and by me in 1993. Collected thalli were dried for storage. Past experience with this preservation method has shown that the tissue integrity of the thalli is not adversely affected, i.e. no cells appear to lyse. In the lab at U.B.C. thalli were rehydrated for fifteen minutes in sea water.

For each sampling time, in 1990/91, twelve thalli were randomly selected from both the reproductive cystocarpic gametophytes and the reproductive tetrasporophytes. In the cases where there were less than twelve reproductive thalli to choose from all of the thalli were used. In 1993 5 thalli were chosen from among those sampled at each time. In some cases less than 5 thalli were available, consequently at some sampling times as few as 3 thalli represent a phase.

Using a dissecting microscope the number of tetrasporangial sori or cystocarps were counted in a field of view with an area of 1 $cm^2$. Counts of the reproductive bumps were taken from the center of the blade half way between the apophasis and blade tip. Only cystocarps or sori which contained spores have been used in the analysis. The absence of pigmentation was taken as an indicator of spore absence. In order to randomly select reproductive bumps from the blades each blade was divided into a grid with 1cm subdivisions. A point on the grid was chosen using two random numbers and the nearest bump which met the selection criteria was excised. Individual sori and cystocarps were excised from the blades by pushing the small end of a glass pipette through the thallus. In order to keep from excising parts of several sori or cystocarps a bump was required to be at least a millimeter away from its neighbours. The other criterion for selecting a sorus or cystocarp was that it be mature, i.e. dark red, but without a developed release pore.

In order to see all the spores at once the small excised portions of thallus were broken down by heating them in a 10% solution, by weight, of Sodium Hydroxide for approximately 45 minutes. The time required to soften the thallus pieces was dependent on the thickness of the thallus and the temperature of the water bath. Test tubes each containing the excised portions from a single blade were filled with the Sodium Hydroxide solution and placed in a water bath at 80 degrees Celsius. Sodium Hydroxide breaks down the cell walls and connections between the cells while leaving the cell membranes intact. When the thallus portions became soft they were pipetted onto regular microscope slides. A drop of water solution containing 20% karo and 5% formalin was then placed on the slides. By gently pushing a cover slip onto the thallus pieces the spores were exposed in a circular pattern, one to three layers thick. In most cases the spores could be fairly easily counted by focusing up and down at 400X magnification using a compound microscope. In the tetrasporangial sori it was actually the tetrasporangia which were counted. Each sporangium was subdivided into four spores with the divisions usually visible. A portion of each squashed sorus or cystocarp was counted and the total number of spores estimated. Early sub-sampling showed that the squashed preparation of each sorus or cystocarp was fairly homogeneous, so a single sub-sample was used to estimate total spore number. A single sorus or cystocarp was counted from each thallus in the 1990/91 collection time and four samples were taken from each blade in the 1993 collection time. The mean of the four 1993 samples was used in the analysis, after Hurlburt (1984).

The data analysis for comparing the phases within one sampling time was done in the Stats module of Systat (Wilkinson 1990) on an IBM PC clone. The phases were compared using t-tests within each sampling date. Both the number of spores produced per sorus or cystocarp and the mean number of spores produced per $cm^2$ were used to compare the phases. The mean number of spores produced per area was a combination of the mean number of spores produced per reproductive structure and the mean number of reproductive structures per $cm^2$.

The comparisons between sampling times within each phase were done using the MGLH module of systat (Wilkinson 1990). A Bonferroni multiple comparisons test was done to compare within one phase the mean numbers of spores per sorus or cystocarp from each of the sampling times. For the 1993 data there were three sampling times for each phase. In 1990/91 three sampling times were used for the cystocarpic thalli and four for the tetrasporic ones.

As a result of using the same data for several statistical tests the chosen value of alpha (0.05) was divided by the number of tests done, for that reason alpha was either 0.013, 0.017 or 0.025.

## Results

Observations were made on three tetrasporangial sori and three cystocarps from the 1993 sampling time which had lost their pigment presumably due to spore release. In the tetrasporangial sori there were no spores remaining in any of the sori, but in the cystocarps there were many small spore-shaped cells. These "spores" were several times smaller than those which were counted as spores in the mature coloured cystocarps. From the scarcity of spores in the colourless bumps it appears that if a release pore develops most spores are eventually released.

In order to meet the assumption of homogeneity of variance the data were log transformed.

Question 1:.Do Gametophytes and Sporophytes produce comparable numbers of spores per blade?

Table 2.1: The results of the t-tests between the phases at each sampling time comparing the mean number of spores per blade.

| Date | Phase | Spores/Blade | p |
|---|---|---|---|
| May '90 | Cyst. | $2.1\times10^{10}$ | .637 |
| | Tet. | $3.5\times10^{10}$ | |
| Aug. '90 | Cyst. | $1.4\times10^{9}$ | .007 |
| | Tet. | $1.6\times10^{10}$ | |
| Nov. '90 | Cyst. | $3.2\times10^{7}$ | .127 |
| | Tet. | $9.6\times10^{9}$ | |

Where Cyst.= Cystocarpic blades and Tet.= Tetrasporic blades.

Alpha = 0.013.

Only in the August 1990 sampling time was there a significant difference between the number of carpospores and tetraspores found on the blades. In February 1991, no carposporic blades were available so only counts of tetraspores are given.

Figure 2.1: The mean number of spores per bump, and standard error, for samples taken in May, August, and November 1990 for both cystocarpic and tetrasporic blades, and for samples taken in February 1991 for tetrasporic blades only.

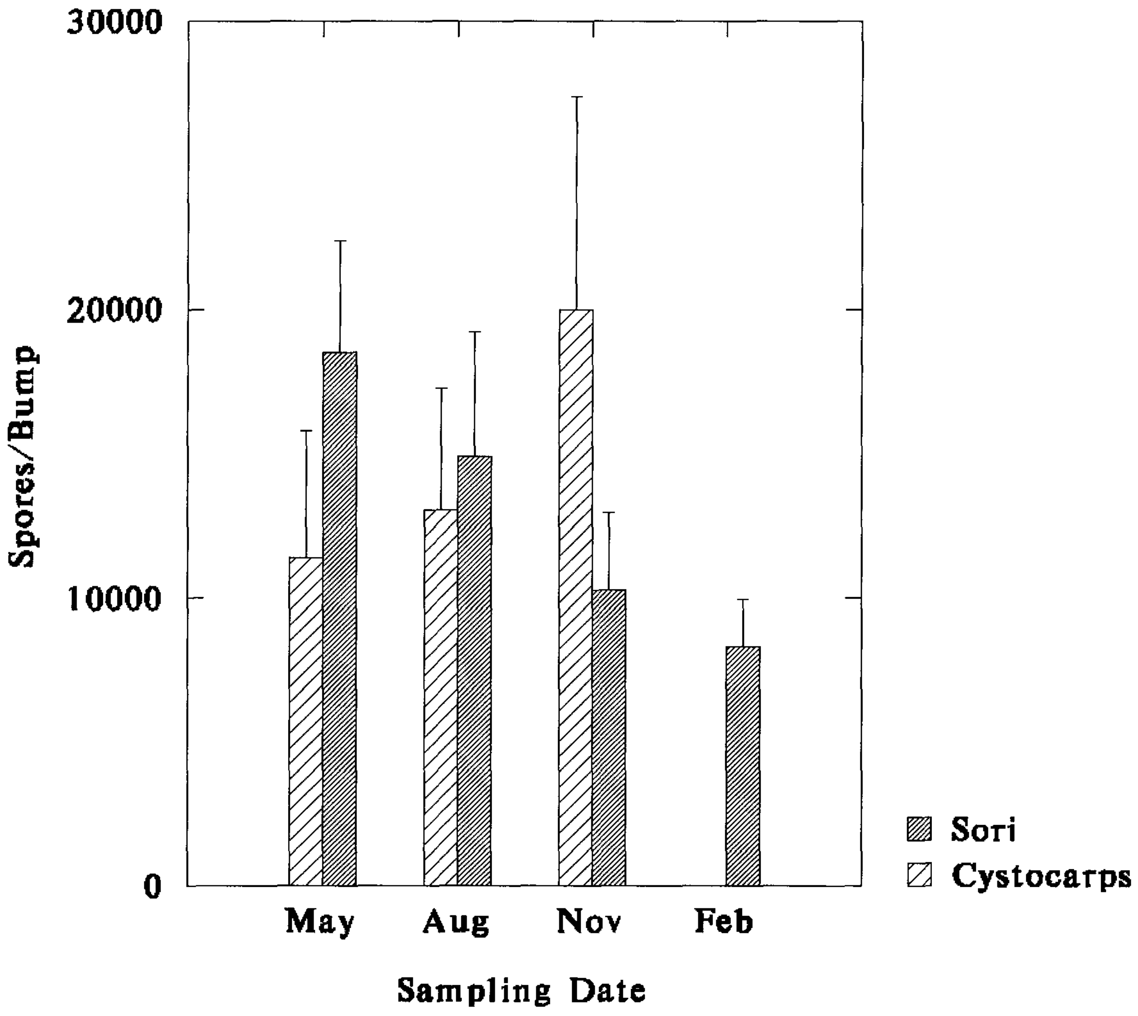


Figure 2.2: The mean number of reproductive sori or cystocarps per cm$^2$, and standard errror, for cystocarpic and tetrasporic blades in May, August, and November 1990, and for tetrasporic blades in February 1991.

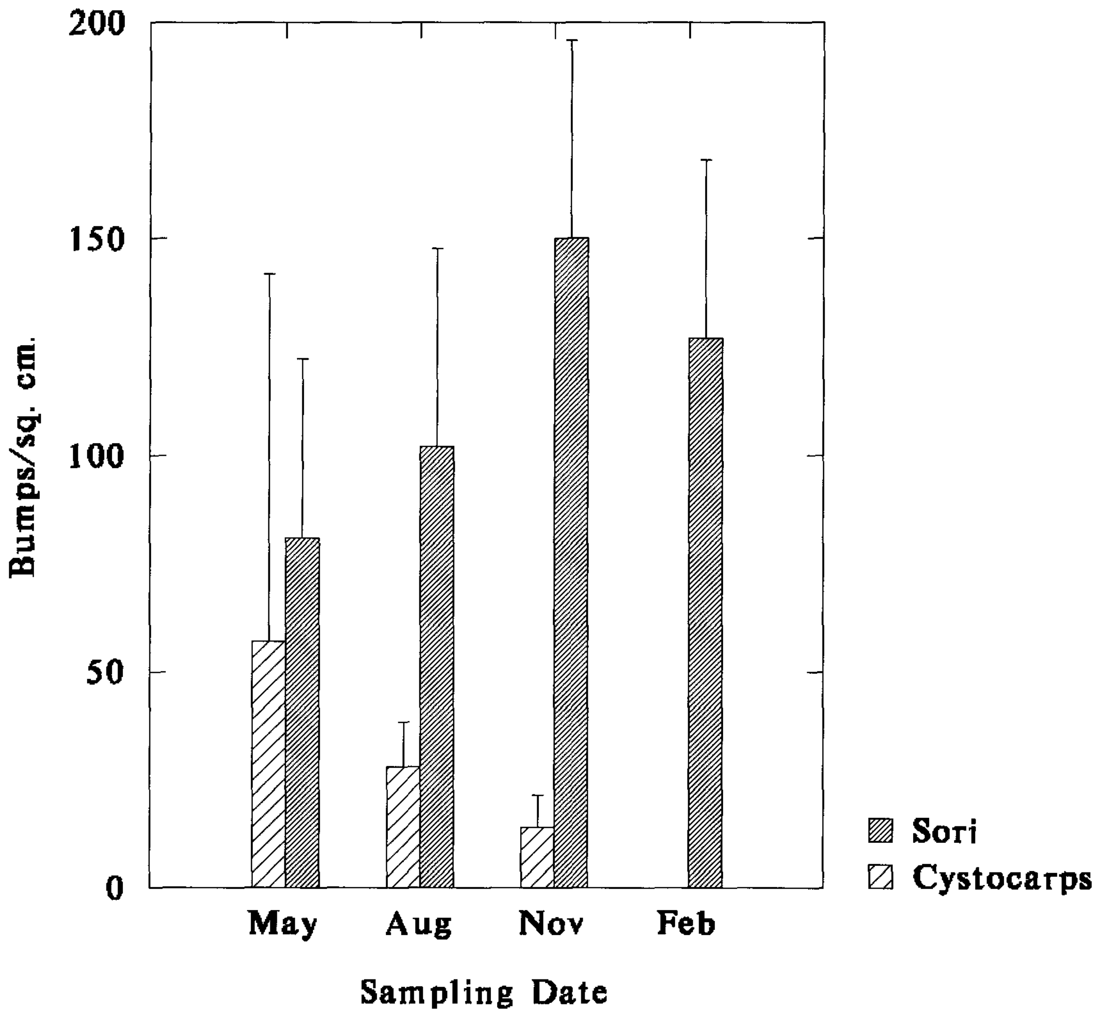


Table 2.2: The mean reproductive blade area, for cystocarpic and tetrasporic blades sampled in May, June, August, and November 1990, and February 1991. The p value for each comparison between the phases is included.

| Date | Phase | Blade Area(cm$^2$) | p |
|---|---|---|---|
| May '90 | Cyst. | 122.7 | .957 |
| | Tet. | 124.6 | |
| June '90 | Cyst. | 80.2 | |
| Aug. '90 | Cyst. | 59.1 | 0.005 |
| | Tet. | 95.1 | |
| Nov. '90 | Cyst. | 6.86 | 0.001 |
| | Tet. | 33.3 | |
| Feb. '91 | Tet. | 8.69 | |

Where Cyst.= Cystocarpic blades and Tet.= Tetrasporic blades.

Alpha = 0.013.

In August and November 1990 the mean reproductive blade area was significantly larger for tetrasporic blades than for cystocarpic blades. In both phases there was trend towards a reduction in reproductive blade area beteen May and November or February.

Question 2:.Is there an annual peak in the production of spores per reproductive structure?

Table 2.3: Multiple comparisons of spores per cystocarp from samples taken in May, June, August, and November 1990.

| Comparison | Dates | Means | p |
|---|---|---|---|
| spores/cyst. | May vs. Aug. | 11400 vs. 13000 | 1.000 |
| | May vs. Nov. | 11400 vs. 20000 | .212 |
| | Aug. vs. Nov. | 13000 vs. 20000 | .144 |

Alpha = 0.017.

There were no significant differences found between the numbers of spores counted per cystocarp in any of the months, although there was a trend towards more spores per cystocarps from May to November.

Table 2.4: Multiple comparisons of spores per sorus from samples taken in May, August, and November 1990 and February 1991.

| Comparison | Dates | Means | p |
|---|---|---|---|
| spores/sorus | May vs. Aug. | 18500 vs. 14900 | .543 |
| | May vs. Nov. | 18500 vs. 10300 | .005 |
| | May vs. Feb. | 18500 vs. 8300 | .000 |
| | Aug. vs. Nov. | 14900 vs. 10300 | .370 |
| | Aug. vs. Feb. | 14900 vs. 8300 | .016 |
| | Nov. vs. Feb. | 10300 vs. 8300 | 1.000 |

Alpha = 0.013.

There were significant differences in the number of spores per sorus between May and November, May and February, and August and February. There was a general trend towards a decrease in the number of spores per sorus as the thallus aged.

Table 2.5: Multiple comparisons of spores per cystocarp from samples taken in February, June, and October 1993.

| Comparison | Dates | Means | p |
|---|---|---|---|
| spores/cyst. | June vs. Oct. | 6300 vs. 12800 | .297 |
| | Feb. vs. June | 24700 vs. 6300 | .003 |
| | Feb. vs. Oct. | 24700 vs. 12800 | .050 |

Alpha = 0.025.

There were significant differences in the numbers of spores per cystocarp between February and June, and February and October, with generally increasing numbers of spores per cystocarp as the blade ages.

## Discussion

Only in August was there a significant difference between the numbers of spores produced on cystocarpic and tetrasporic blades (Table 2.1), with more spores being produced by the tetrasporophytes. The significant difference in the number of spores produced per blade in August was a combination of the greater number of reproductive structures per $cm^2$ (Figure 2.2) and the greater blade size (Table 2.2). The lack of significant differences in the number of spores per blade at other times was the result of the interactions between the number of spores per bump, bump density, and blade size.

Not only were as many or more tetraspores found per blade as carpospores, at all sampling times, but the number of reproductive tetrasporic blades outnumbers the number of cystocarpic blades by several times throughout the year in the study population (Shaughnessy Pers. Comm.). Although the peak in cystocarpic blades in the sampled population occurs in the late spring and summer the reproductive tetrasporophytes still outnumber them by several times. How then is the population of tetrasporophytes maintained at such a high level with tetraspores forming the vast majority of spores produced?

The answer may be found in recruitment (Chapter 3), perhaps in spite of the high tetraspore production more tetrasporophytes recruit than gametophytes. Alternatively, the mortality rate of gametophytes may be much higher than that of the tetrasporophytes. One potential mechanism, wave induced mortality, is discussed in Chapter 4. Another potential cause of phase biased mortality is herbivory. Luxoro and Santelices (1989) have shown that some intertidal snails prefer one phase over the other. In Iridaea laminarioides the preference is for tetrasporophyte tissue. Furthermore, Buschmann (1991) demonstrated that the amphipod Hyale hirtipalma has a strong preference for cystocarpic tissue. It has not been demonstrated that feeding preferences significantly affect the growth of either the gametophytes or tetrasporophytes at the population level.

As the gametophyte blades matured the number of spores per cystocarp appeared to increase (Tables 2.3 and 2.5). The opposite was true for the tetrasporophytes: as the blades matured the number of spores per sorus decreased (Table 2.4). The lowest numbers of spores per bump were found during the period of highest reproductive blade density for each phase. If there are significant costs associated with reproduction the low number of spores per bump may correspond to higher growth rates. It has not been demonstrated for the red algae that there are costs associated with reproduction since the spores are pigmented and may support their own growth through photosynthesis (DeWreede and Klinger 1988). Growth rates for the gametophytes and tetrasporophytes of Mazzaella lilacina have not been compared during the necessary periods so any connection between growth rate and spore production is purely speculative.

Another part of the puzzle is the relationship between the number of spores per bump and the density of bumps. Figures 2.1 and 2.2 suggest that the two may be interdependent. In the tetrasporophytes (Figure 2.1) there was a progressive decrease in the number of spores per sorus while at the same time (Figure 2.2) there was a progressive increase in the density of sori. The opposite was true for the gametophytes. If the production of spores entails significant costs to the thallus then an increase in the number of spores per bump would be expected to be associated with a decrease in the number of bumps, or a decrease in the growth rate. The resolution of the above possibility is beyond the scope of this study since some of the trends mentioned in Figures 2.1 and 2.2 were not statistically significant and even if they were, no causal relationship could be established.

One point which has been made abundantly clear by this work is that the success rate for spores of Mazzaella lilacina is very low. In May of 1990 the average number of spores carried by cystocarpic and tetrasporic blades was 21 and 25 billion, respectively. Many empty cystocarps and sori are found on the blades at this time suggesting that a large number of spores have already been released. Add to this that only about 20% of the population each year is recruited from spores (May 1986 for Iridaea cordata) and the successful recruitment rate becomes as low or lower than 1 in 175 billion.

It has been demonstrated that the lowest number of spores found in sori and cystocarps was during the period of peak reproductive blade density for each phase. As well, the production of tetraspores was much greater than the production of carpospores at all times of year. The following chapter will investigate the recruitment of gametophytes and tetrasporophytes into both a wave exposed environment and a more sheltered one.

# CHAPTER 3: RECRUITMENT OF *Mazzaella lilacina*

## Introduction

Three models, which have been proposed to account for distributions of the gametophytes and tetrasporophytes of isomorphic algae, will be tested in this chapter.

Lazo et al. (1989) proposed a model for areas with stable substrata, in which the competitively dominant phase would become numerically more abundant than the subordinate phase. Dominance would be maintained by vegetative regeneration of blades from the perennial holdfasts, with little recruitment from spores. Their research focused on an isomorphic member of the Gigartinales, Chondrus crispus Stackhouse, on the east and west coasts of Prince Edward Island, Canada. The stable substratum model requires that there be differential mortality among phases due to competition or other forces, and assumes that recruitment from spores is not a major factor. Lazo et al. (1989) cite work by Bhattacharya (1985). In southwestern Nova Scotia, Canada, a population of C. crispus on stable substrate had nearly 100% cover and was composed of 90% gametophytes. If regeneration from the holdfast was the primary means of producing new blades, a commonly reported phenomenon among red algae (DeWreede and Green 1990, D'Antonio 1986, May 1986, Hansen and Doyle 1976), then in spite of the high output of tetrasporophyte producing carpospores, gametophyte dominance could persist indefinitely.

At my research sites Mazzaella lilacina, unlike Chondrus crispus, did not grow in very dense patches; moreover, there was little if any overlap of blades from neighbouring holdfasts. Thus, it seems unlikely that competition between the phases occurs to any significant extent.

Other factors which may cause differential mortality among the phases are: 1) age -- one of the phases may have a much lower maximium biological age, a possibility addressed by Shaughnessy (Pers. Comm.), 2) herbivory -- Luxoro et al. (1989) and Buschmann et al. (1993) both report phase preferences by invertebrates for Iridaea laminarioides , and 3) removal by waves -- Dyck et al. (1985) found that the proportion of gametophytes of Mazzaella lilacina (as Iridaea cordata) in the drift was higher than the proportion found in the intertidal population, suggesting that gametophytes were more easily removed than tetrasporophytes. Differences in susceptibility to wave induced removal of blades will be addressed in Chapter 4.

In the model proposed by Lazo et al. (1989) for stable substrata the final phase proportions of a population are due to an equilibrium between recruitment and mortality resulting from competition or some other process which favours one phase over the other. If this model is not appropriate then the proportions of the phases at the time of recruitment will remain constant over an extended period. Changes in favour of one phase would suggest that phase discriminating mortality is occurring, supporting the model by Lazo et al. (1989). If the proportions of gametophytes and tetrasporophytes are constant over time then either no phase discriminating sources of mortality were at work or that the active forces counteracted each other. The first question of this study addresses whether or not the model proposed by Lazo et al. (1989) for stable environments is appropriate for my study sites. Do the proportions of gametophytes and tetrasporophytes in the populations of new recruits change in favour of one phase?

If the environment is more disturbed several researchers propose a stochastic model (Dyck et al. 1985, Lazo et al. 1989, May 1986) in which phase recruitment proportions following a disturbance are determined by the proportions of available spores. In a purely stochastic model following recruitment there would be phase differentiating forces which would act to change the phase proportions of the population. Patch dominance would depend entirely on which spores were most abundant at the time of the disturbance. In the work by Lazo et al. (1989) the overall population of Chondrus crispus on P.E.I during the two summers of sampling had a phase ratio of nearly 1:1, with gametophytes comprising 56% of the population. Lazo et al. (1989) indicated that this ratio was consistent with the stochastic model which they proposed. An expected 1:1 ratio assumes that the gametophytes and tetrasporophytes are identical ecologically, a premise which has not been supported in the literature

Using the stochastic model one would expect the proportions of gametophytes and tetrasporophytes among recruits to reflect the proportions of each phase among available spores. Judging from the natural population in the exposed site (Shaughnessy Pers. Comm.), at some point more tetrasporophytes than gametophytes must recruit; the opposite must be true for the sheltered site. However, if gametophytes always recruit in greater numbers than tetrasporophytes in the exposed site, and tetrasporophytes always recruit in greater numbers in the sheltered site, then a stochastic model is not appropriate. The second question to be examined in this chapter follows the test outlined above;

do more gametophytes recruit than tetrasporophytes regardless of the timing of clearing in the exposed site, and do more tetrasporophytes than gametophytes recruit regardless of the timing of clearings in the sheltered site?

The model for stable environments and the stochastic model are not mutually exclusive. For example, immediately following settlement the proportions of gametophytes and tetrasporophytes may reflect spore availability, however, as the thalli mature competition or other phase differentiating processes may become important.

Another model, based on observations made on Mazzaella lilacina, has been proposed by Shaughnessy (Pers. Comm.). This model combines parts of both of the above models. In the "Nobleweed-Pauperweed" model Shaughnessy postulates that the gametophytes are specialized for recruitment while the tetrasporophytes are specialized to live longer and produce more spores than the gametophytes. While, the mechanism behind recruitment specialization as applied to the gametophytes will not be examined here, some possibilities are 1) tetrasporophytes produce more spores, or 2) tetraspores are able to germinate in a wider range of microhabitats. If the tetrasporophytes are longer lived it may be the result of having a greater resistance to various sorts of disturbance, not necessarily a greater maximum biological age.

If this model is appropriate for my sites then regardless of the time of year at which clearings were made or their placement within the Mazzaella lilacina zone there would be a greater proportion of gametophyte recruits than the proportion of tetrasporophytes in the surrounding 'parent' population. The parent population refers to the reproductively mature thalli at the time clearings were made. It is not sufficient to have more gametophyte recruits than tetrasporophytes because in cases where the surrounding population has many more tetrasporophytes than gametophytes, even if the phases were ecologically identical one would expect more gametophyte recruits than tetrasporophytes, so finding that result would not indicate any sort of 'specialization'. Based on the above expectations a third question will be addressed in this chapter; is the proportion of gametophyte recruits higher than the proportion of reproductive tetrasporophytes in the surrounding population at the time of the disturbance?

The second tenet of the 'Nobleweed-Pauperweed' model is that tetrasporophytes are longer lived. Therefore, one would expect that the proportion of tetrasporophytes in the clearing populations would increase over time. The fourth question to be addressed is, does the proportion of tetrasporophytes among the recruits increase over time?

This study doesn't separate the potential mechanisms underlying differences in recruitment it only tests to see if there were differences in the recruitment of juveniles (>.5cm) of each phase at different times of year and in the two sites.

Finally, statistical tests were done to test what appeared to be obvious in the field, namely, that the proportion of tetrasporophytes was greater in the more wave exposed study area than in the sheltered area.

In summary; the null hypotheses to be tested in this chapter are:
1) The proportions of gametophytes and tetrasporophytes following recruitment do not change with time.

2) In the exposed site as many or fewer gametophytes than tetrasporophytes will recruit regardless of when clearings are made; in the sheltered site as many or fewer tetrasporophytes than gametophytes will recruit regardless of when clearings are made.

3) The proportion of tetrasporophyte recruited into a clearing was greater than or equal to the proportion of reproductive gametophytes in the surrounding population, at the time the clearing was created.

4) The proportion of tetrasporophytes found in the clearings decreased over time.

5) In the more wave exposed area the population of Mazzaella lilacina had a smaller or equal proportion of tetrasporophytes as the population in the less wave exposed area.

## Materials and Methods

Field sampling in the sheltered and exposed sites was carried out during periods of low tide at approximately two month intervals from November 1991 to October 1993.

Within each site (sheltered and exposed) several transects were haphazardly placed within the Mazzaella lilacina zone. The sheltered site contained seven transects and the exposed site four. Both ends of the transects were marked by a concrete nail pounded into the rock, with a piece of brightly colored flagging tape tied to the nail. Evenly spaced along each transect were nine 25cm by 25cm quadrats. Neighboring quadrats were placed on opposite sides of the transect line and separated horizontally by a quadrat width. Six randomly chosen quadrats on each line were used for two replicates of each of three clearing times. Three quadrats on each line were used for replicates of uncleared controls. Quadrat sized clearings were made using an air powered chisel, in November of 1991, at the peak of reproductive tetrasporophyte abundance, in early May of 1992, during the peak of vegetative abundance, and in July 1992, at the peak of reproductive gametophyte abundance. All living organisms were removed in the cleared areas along with some of the underlying rock. For each clearing time, in the sheltered site, there were 14 replicate clearings, and a total of 21 control quadrats. In the exposed site there were 8 clearings for each of the three clearing times, and a total of 11 controls.

Control quadrats were used to monitor the surrounding population for phase abundance and reproductive timing. Treatments and controls were sampled several times during 1991,1992, and 1993. Table 3.1 shows sampling times as well as clearing times.

Table 3.1: The clearing and sampling schedule for both the sheltered and exposed sites. Where, C refers to sampling of the controls, T1 refers to sampling of the November 1991 clearings, T2 for the May 1992 clearings, and T3 for the July 1992 clearings.

| Sampling Time | Clearings | Sheltered Site | Exposed Site |
|---|---|---|---|
| 11/91 | T1 | C | C |
| 02/92 | | C,T1 | C,T1 |
| 04/92 | | C,T1 | |
| 05/92 | T2 | C,T1 | C |
| 07/92 | T3 | C,T1 | C,T2 |
| 10/92 | | C,T1,T2 | C,T1,T2,T3 |
| 02/93 | | T1 | T1,T2,T3 |
| 04/93 | | T1,T2,T3 | |
| 06/93 | | T1,T2,T3 | T1,T2,T3 |
| 10/93 | | T1,T2,T3 | T1,T2,T3 |

The largest blade from each holdfast in the clearings and controls was measured for length and, if reproductive, its phase was recorded. Blades smaller than .5cm in length were not counted because of difficulties in differentiating similar taxa. If a thallus was not reproductive then a small portion, about .2 $cm^2$, of the blade was taken and identified in the lab using the resorcinol/acetal test (Garbary and DeWreede 1988).

In addition to counting blades of Mazzaella lilacina, recruitment of all invertebrate and algal taxa was measured as percent cover and used to compare the biotic component of the sheltered and exposed sites (Chapter 1).

## Data Analysis

In order to compare the abundances of gametophytes and tetrasporophytes the mean proportions, and 95% confidence limits, for the phases within each site were calculated using the 'Tables' module of "Systat" (Wilkinson 1990). In order to have a sufficient number of recruits (25+) over 0.5cm long the interval between creation of a set of clearings and analysis of the recruitment data varied. For example, in the sheltered site the first sampling time with enough recruitment for analysis after clearing time 1 (November 1991) was in April of 1992 (5 months), whereas for clearing time 2 the time lapse was 11 months (May 1993 to April 1993).

Tests on independent proportions (Hicks 1982) were used to determine if the proportion of gametophytes in the recruits and the proportion of reproductive tetrasporophytes in the surrounding population at the time the clearings were made were the same. Because the number of reproductive tetrasporophytes in the control quadrats was too small, in the sheltered site the proportions of gametophyte recruits from the time 2 (May 1992) and time 3 (July 1992) clearings were each compared to a sample of the reproductive tetrasporophyte population taken in June 1993. In the exposed site, the time 1 (May 1992) and the time 2 (July 1992) gametophyte recruit proportions were each compared to the proportion of reproductive tetrasporophytes sampled in July 1992.

The reproductive blade samples used above were collected using a haphazardly placed transect line with randomly spaced marks on it. The nearest reproductive blade to each mark was collected, and the overall phase proportions for reproductive blades in both the sheltered and exposed sites were calculated. Recruits into clearings made in November 1991 (time 1) in the exposed site were compared to samples taken from the controls at the time the clearings were made. No comparisons were made for the recruits into the November 1991 (time 1) clearings in the sheltered site. Table 3.2 in the results section shows all of the comparisons made between recruits and 'parents'.

In order to determine whether or not there was a significantly greater proportion of tetrasporophytes in the exposed site than in the sheltered site significance tests on independent proportions were used (Hicks 1982). The proportions of tetrasporophytes in the exposed site control quadrats were compared to the proportions of tetrasporophytes in the sheltered site control quadrats at three different times, November 1991, May 1992, and July 1992. The three comparison times correspond to the three times at which clearings were made, i.e. the peak of tetrasporophyte reproduction, the peak of vegetative blade abundance, and the peak of gametophyte reproduction.

## Results

The data for the first hypothesis are presented in graphical form in Figures 3.1,3.2,and 3.3. Hypothesis 1 states that following recruitment the proportions of the phases will not change as the cohort ages. If this hypothesis is supported then the model proposed by Lazo et al

(1989) for stable substratum was not appropriate in the clearings. The hypothesis is non-directional, in order to reject the hypothesis it only matters that the proportions changed in favour of one of the phases.

Figure 3.1: Mean gametophyte abundance measured as a percentage of the total number of recruits into clearings made in November 1991 in the sheltered site. 95% confidence limits surround each measurement.

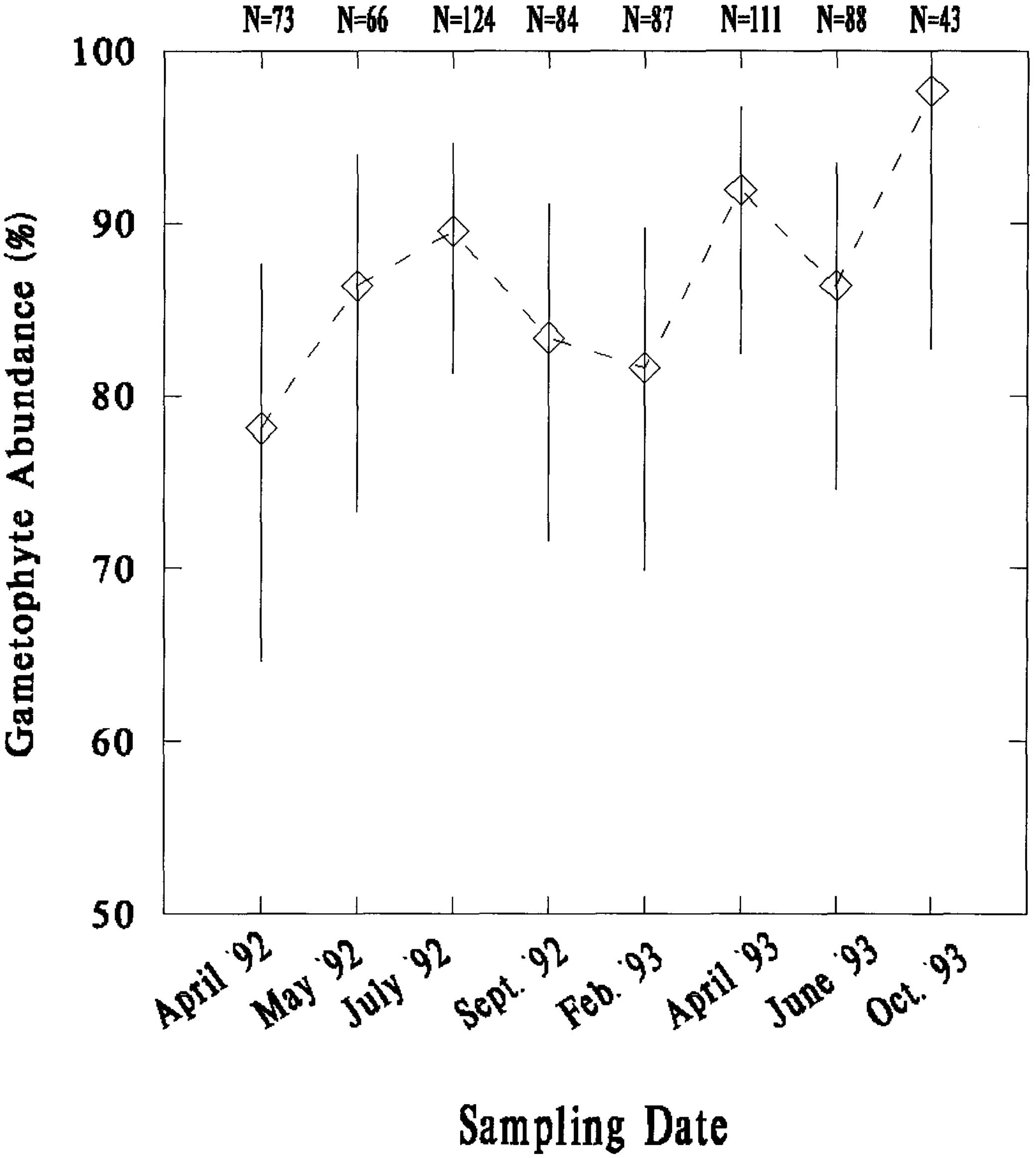


Figure 3.2: Mean gametophyte abundance measured as a percentage of the total number of recruits into clearings made in November 1991 in the exposed site. 95% confidence limits surround each measurement.

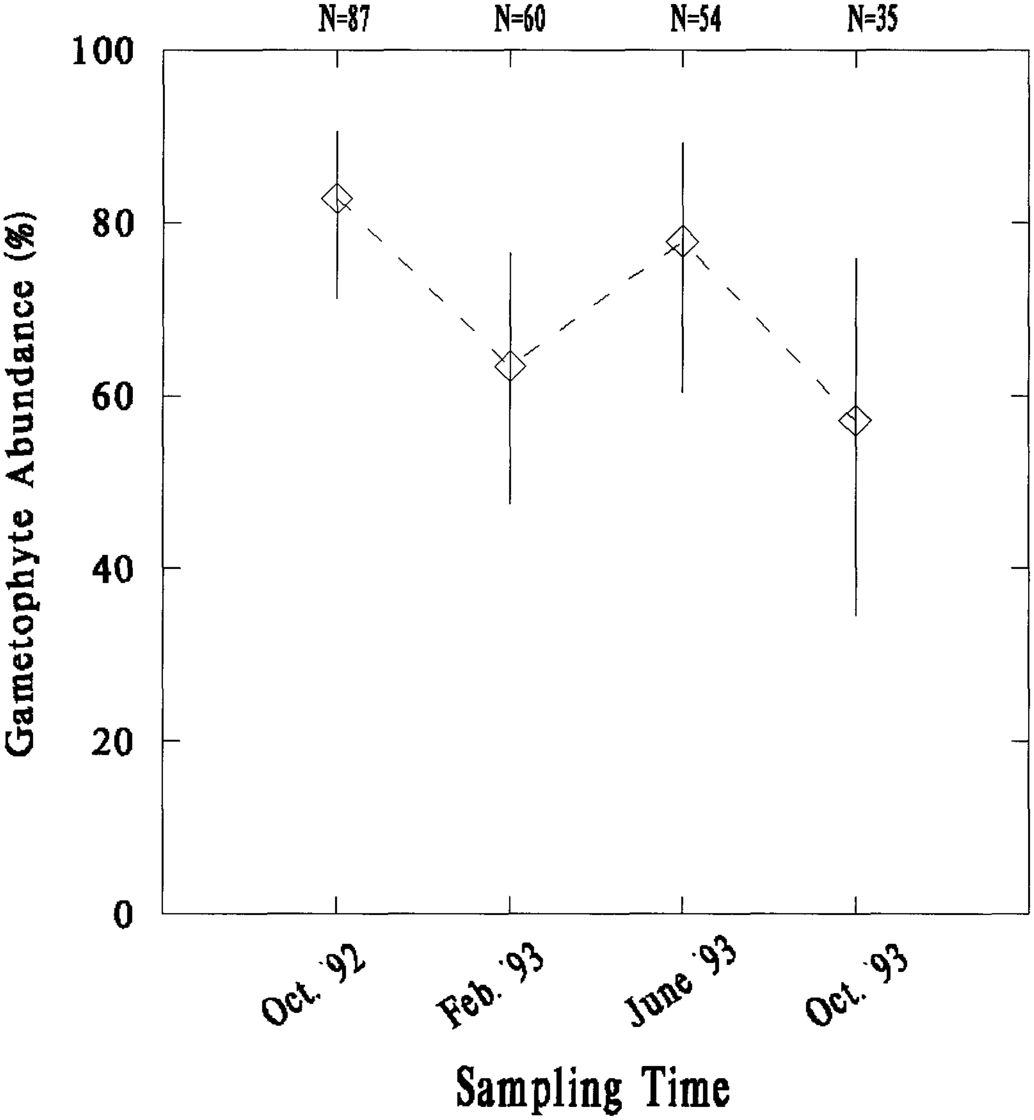
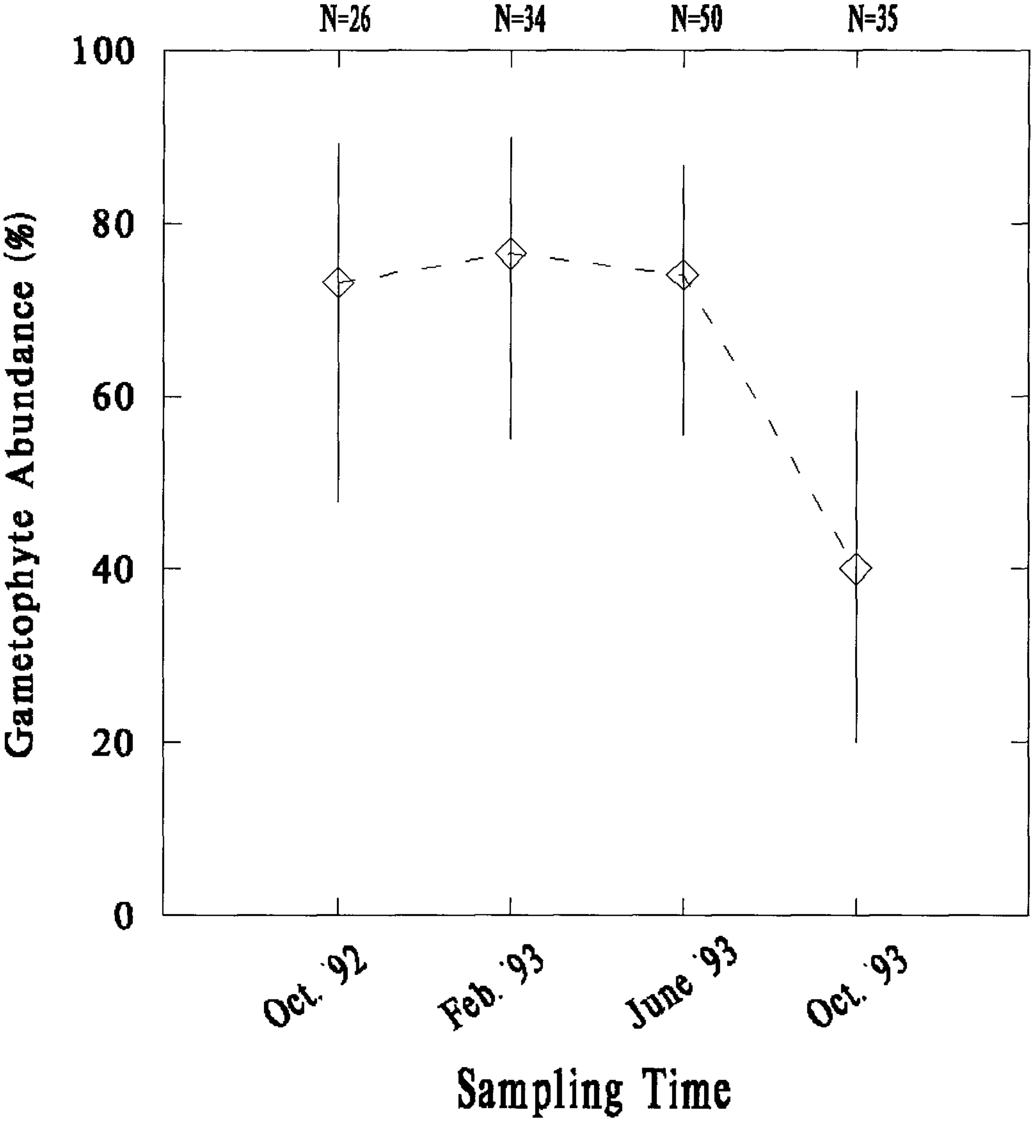


Figure 3.3: Mean gametophyte abundance measured as a percentage of the total number of recruits into clearings made in May 1992 in the exposed site. 95% confidence limits surround each measurement.



The above graphs do not show a statistically significant change in the phase proportions in the repeated sampling following recruitment. There do appear to be trends, however; the proportion of gametophytes appeared to increase in the sheltered site (Figure 3.1) and decrease in the exposed site (Figures 3.2 and 3.3).

Hypothesis 2 is a test of the stochastic model and states: in the exposed site an equal number or fewer gametophytes will recruit than tetrasporophytes regardless of when clearings were made; in the sheltered site an equal number or fewer tetrasporophytes will recruit than gametophytes regardless of when clearings were made. If the hypotheses are rejected then the stochastic model is not appropriate for explaining the observed phase proportions in either the sheltered or exposed site. The analysis of hypothesis 2 is presented in graphical form (Fig. 4 and 5). For each clearing time the first samples of recruitment in each site when more than 25 blades were found are presented. Bars represent the mean percent of recruits which were gametophytes.

Figure 3.4: Mean gametophyte abundance in the sheltered site, measured as a percentage of the total recruitment, in the first sampling time (N=25+) following each clearing event. The date above each bar was when sampling occurred. Each bar is surrounded by 95% confidence limits.

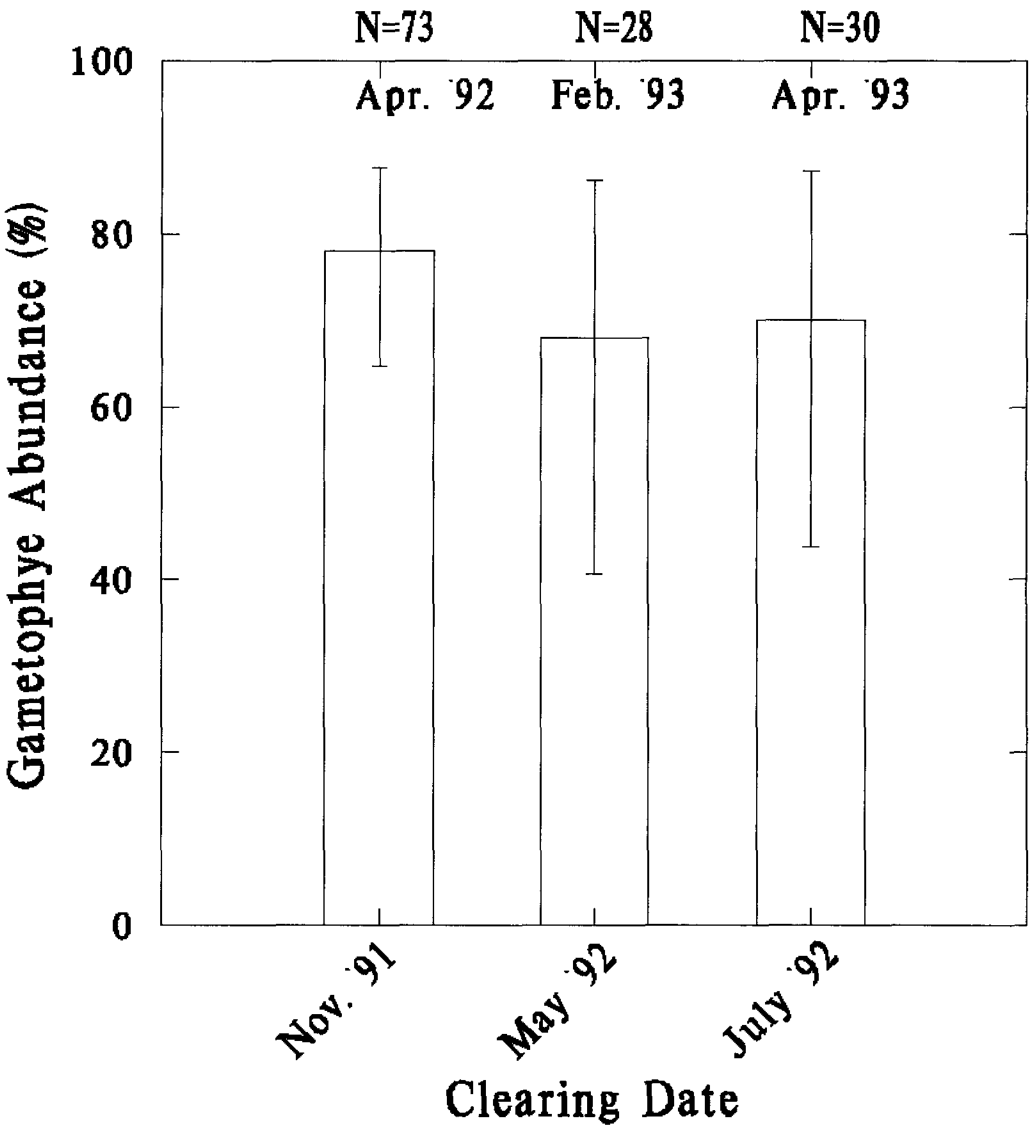
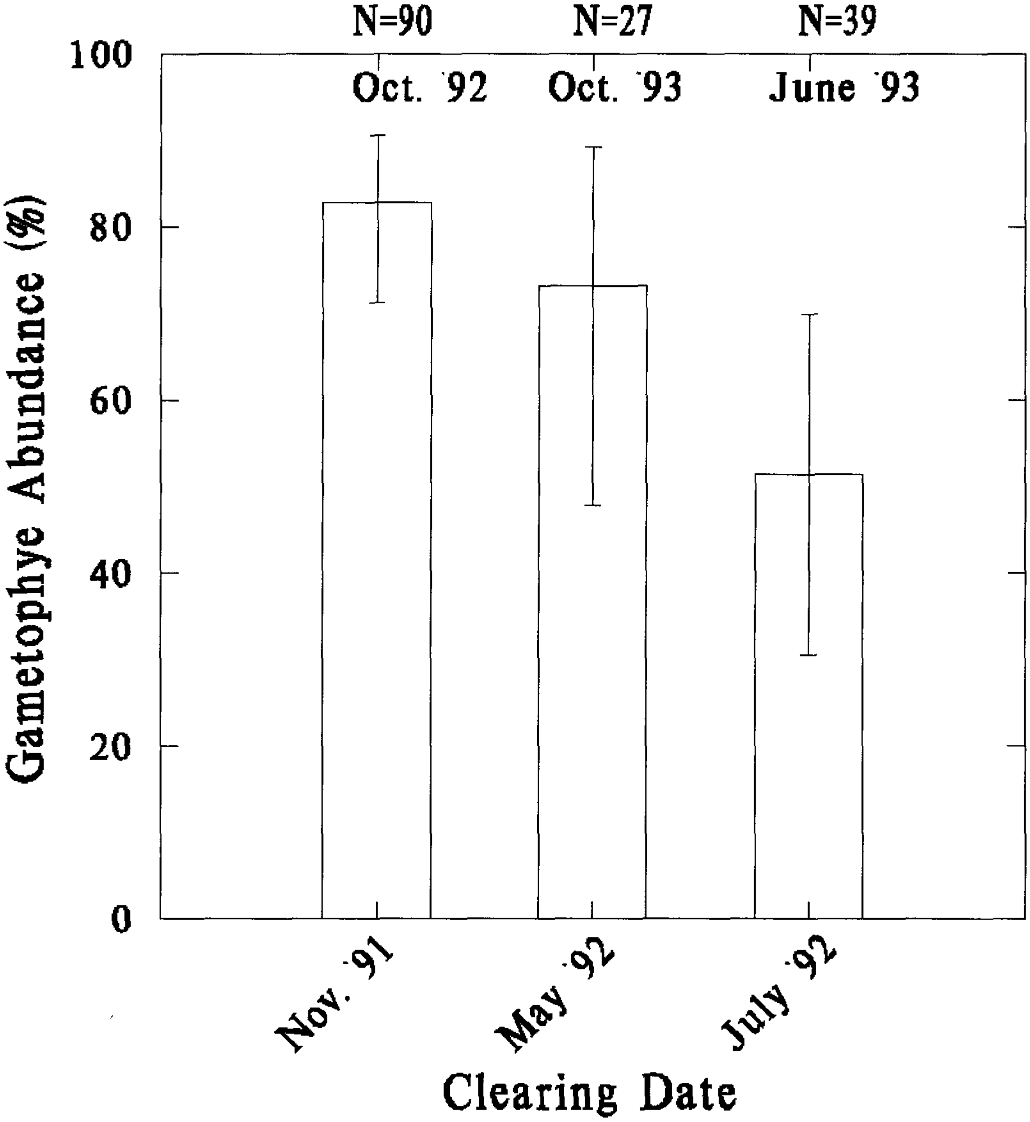


Figure 3.5: Mean gametophyte abundance in the exposed site, measured as a percentage of the total recruitment, in the first sampling time (N=25+) following each clearing event. The date above each bar was when sampling occurred. Each bar is surrounded by 95% confidence limits.



In the sheltered site at all times there were significantly or nearly significantly more gametophyte recruits than tetrasporophytes. The same was true for the exposed site in the clearings made in November 1991 and May 1992, however, in July 1992 the proportions of gametophytes and tetrasporophytes were very close. Hypothesis 2 cannot be rejected for the sheltered site, suggesting than the stochastic model may be appropriate in this site. In the exposed site, while hypothesis 2 cannot be rejected at no time did as great a proportion of tetrasporophytes recruit as the proportion found in the surrounding population, suggesting that the stochastic model is not appropriate.

Hypothesis 3 is a test of the first part of the 'Nobleweed-Pauperweed' model and states that regardless of the time or place of the clearing the proportion of gametophyte recruits will be greater than the proportion of reproductive tetrasporophytes in the population at the time of the clearing. If hypothesis 1 is rejected then the 'N-P' model is not appropriate in the study sites. The data are presented below in Tables 3.2 and 3.3. The critical Z value for all of the tests below is 1.64. Alpha was set at 0.05.

Table 3.2: The proportion of gametophyte recruits vs. the proportion of reproductive tetrasporophytes, i.e. 'Parents', in the sheltered site. Both comparisons use information on the reproductive blade population gathered in June 1993.

| Clearing Time | Recruits | N | 'Parents' | N | Z |
|---|---|---|---|---|---|
| Nov. 1991 | N/A | | | | |
| May 1992 | .68 | 28 | .32 | 37 | 2.83 |
| July 1992 | .70 | 30 | .32 | 37 | 3.06 |

Table 3.3: The proportion of gametophyte recruits vs. the proportion of reproductive tetrasporophytes, i.e. 'Parents', in the exposed site. The November comparison uses reproductive blade population information from the controls in November 1991. The May and July comparisons use information from a census of the reproductive blade population made in July 1992.

| Clearing Time | Recruits | N | 'Parents' | N | Z |
|---|---|---|---|---|---|
| Nov. 1991 | .86 | 83 | .96 | 25 | 1.30 |
| May 1992 | .66 | 29 | .85 | 20 | 1.52 |
| July 1992 | .51 | 39 | .85 | 20 | 2.54 |

In the sheltered site for both clearing times the null hypothesis of no difference between the proportions could be rejected. In the exposed site, only for July 1992 clearings (time 3) was a significant difference in the proportions of gametophyte recruits and tetrasporophyte 'parents' found. However, the difference was in the opposite direction to what was expected. The difference was approaching significance for the November and May clearing times ( significant at alpha = 0.10 and 0.066, respectively) but again in the opposite direction to what was expected.

Hypothesis 4 is a directional version of hypothesis 1 and states: following recruitment the proportion of tetrasporophytes will increase. If the proportions of tetrasporophytes do not increase then the 'Nobleweed-Pauperweed' model was not appropriate to the study sites. Figures 3.1, 3.2, and 3.3 will be used to examine this hypothesis.

None of the graphs showed a statistically significant trend towards increased tetrasporophyte proportions with time following recruitment. However, in both sites slight trends were apparent. The trend in the sheltered site was towards increased gametophyte proportions. In the exposed site the opposite trend was suggested; towards an increased proportion of tetrasporophytes.

This study was based on the premise that the proportions of tetrasporophytes in the more wave exposed site were higher than in the sheltered site. Hypothesis 5 was a test of that premise: the proportion of tetrasporophytes increased with wave exposure. Table 3.4 contains the data from comparisons of the proportions of tetrasporophytes from the sheltered and exposed populations at three times of the year. Alpha was set at 0.05 and the critical value for Z set at 1.64.

Table 3.4: Proportions of tetrasporophytes, comparing the sheltered and exposed sites.

| Time | Sheltered | N | Exposed | N | Z |
|---|---|---|---|---|---|
| Nov. 1991 | .444 | 54 | .789 | 52 | 2.27 |
| May 1992 | .429 | 28 | .765 | 34 | 2.70 |
| July 1992 | .429 | 28 | .712 | 52 | 2.48 |

At all of the sampling times the null hypothesis of no difference between the proportions was rejected. The proportion of tetrasporophytes in the exposed site was consistently higher than the proportion of tetrasporophytes in the sheltered site.

## Discussion

Based on the model for stable substratum used to explain variations in phase abundance for isomorphic algae (Lazo et al. 1989) the proportions of gametophytes and tetrasporophytes were expected to alter in favour of one phase or the other with time following recruitment (Hypothesis 1). This study used two populations subject to very different levels of wave activity and other related conditions such as desiccation. I made no assumptions about which site *Mazzaella lilacina* found more harsh, my only intention was to use the sites as examples of different physical conditions.

In the sheltered site the recruits in the November 1991 (T1) clearings were followed for fourteen months and the phase proportions did not vary significantly during that time (Figure 3.1). The same was true of the November 1991 (T1) and May 1992 (T2) clearings in the exposed site (Figures 3.2 and 3.3). While, no significant changes in proportions occurred, trends were beginning to show. In the sheltered site the trend in the November 1991 clearings was towards an increased proportion of gametophytes; in the exposed site in the November 1991 and May 1992 clearings the trends were towards increased proportions of tetrasporophytes. The implication is that biotic or abiotic factors in the two sites favour different phases. It appears that gametophyte mortality was higher than tetrasporophyte mortality in the exposed site, and vice versa in the sheltered site. Although the trends were not statistically significant, the 'stable model' of Lazo et al. (1989) appears to have been supported under both sets of conditions.

One difference between the experimental set up used in this study and an unmanipulated population is that all of the blades present in the clearings recruited within a short period of time so that most of the recruits were not in close proximity to large algal blades. In natural populations most clearings are probably much smaller, the result of invertebrate feeding, for example. Consequently, there are factors involving the close proximity of mature thalli which may be important *in situ* which cannot be evaluated. Destombe et al. (1993) reported for the red alga Gracilaria verrucosa (Hudson) Papenfuss that the holdfasts of haploid juveniles grew better than those of diploid juveniles under non-optimal conditions.

Hypothesis 2 was used to test the model for disturbed environments, i.e. the stochastic model (Dyck et al. 1985, Lazo et al. 1989, May 1986), where the phase proportions in a population are a reflection of spore availability at the time space becomes available. Significantly or nearly significantly more gametophytes recruited than tetrasporophytes into clearings made in the sheltered site, regardless of when the clearings were made (Figure 3.2). In the natural population in the sheltered site there are also more gametophytes than tetrasporophytes so it appears than a stochastic model may be an appropriate predictor of final phase proportions in that site. However, in the exposed site (Figure 3.5), while the surrounding population is dominated by tetrasporophytes the more gametophytes than tetrasporophytes recruited into clearings made in November 1991 and May 1992. The recruits into the July clearings had the smallest proportion of gametophytes in exposed site clearings, as would be expected since that is the time of the greatest cystocarpic blade density. However, even the recruitment event with the highest proportion of tetrasporophytes, ca. 0.50 in the exposed site clearings, fell short of the proportions found in the surrounding population (ca. 0.70), suggesting that the surrounding population is not merely a reflection of spore availability or recruit proportions following disturbances. Following recruitment more gametophytes must die than tetrasporophytes if the populations in the clearings are to match those in the surrounding areas.

The tenet of the Nobleweed-Pauperweed model that gametophytes are 'specialized' for recruitment (Hypothesis 3) was supported for the sheltered site (Table 3.2). Specialization may be as simple as greater spore production per tetrasporic blade than per gametophytic blade as was shown in Chapter 2, or a greater ability to succeed in the site.

The gametophytes recruited in greater proportion than the proportion of reproductive tetrasporophytes at the time the clearings were made.

The results in Table 3.3 showed something quite different than what was found in the sheltered site. In the exposed site the gametophytes recruited in a significantly smaller proportion than the proportion of reproductive tetrasporophytes present in the untreated population at the time clearings were made. According to these data the tetrasporophytes were specialized for recruitment. Looking at the results from the sheltered site and the exposed site together it appears that for some reason the gametophytes were favoured in the sheltered site and tetrasporophytes were favoured in the exposed site. Environmental factors may influence the success rate of the spores or sporelings: alternatively, the phases might produce different amounts of spores in the two sites (only the exposed site was looked at in Chapter 2).

The Nobleweed-Pauperweed model relies on gametophytes possessing some sort of specialization for recruitment. Based on the above analysis, the data do not support the idea that gametophytes are always better recruiters. However, it should be noted that in almost every case (Figures 3.4 and 3.5) gametophytes recruited in significantly or nearly significantly greater proportion than tetrasporophytes.

How can this apparent disparity in the data be interpreted? While there were more gametophytes recruiting than tetrasporophytes at most times in both sites, in the exposed site the proportion of tetrasporophytes in the reproductive blade population was also very high, thus, in this site even though not as many gametophytes recruited as the proportion of reproductive tetrasporophytes present in the surrounding population at the time the clearings were made, the proportion of gametophyte recruits was higher than the proportion of tetrasporophyte recruits. In both sites the proportion of gametophytes among the recruits must decrease significantly with time if the recruits are to conform to the observed (control) populations. So far no statistically significant change has occurred, but the proportion of gametophytes may actually be increasing in the sheltered site.

The Nobleweed-Pauperweed model has two basic tenets; 1) that the gametophytes are 'specialized' for recruitment, and 2) that the tetrasporophytes are longer lived and thus better able to send out spores (Hypothesis 4). The second tenet, like the first, was not supported unequivocally by this study. Significant trends in the proportions of gametophytes and tetrasporophytes over time were not observed (Figures 3.1, 3.2, and 3.3). The slight trends that appeared suggest that in the sheltered site the mortality of tetrasporophytes is actually higher than the mortality of gametophytes. The opposite appears to have been true in the exposed site. In neither site were both tenets of the Nobleweed-Pauperweed model supported.

The fifth hypothesis was intended to demonstrate statistically what was obvious in the field, that the proportion of tetrasporophytes was higher in the exposed site than in the sheltered site. Table 3.4 showed that at all the initial clearing times the proportion of tetrasporophytes in the exposed site was significantly higher than the proportion of tetrasporophytes in the sheltered site.

In summary, 1) contrary to the stable substratum model (Lazo et al. 1989) competition or other forces causing differential mortality between gametophytes and tetrasporophytes did not cause statistically significant changes to the proportions of gametophytes and tetrasporophytes in the clearings, in either site. However, there did appear to be a slight trend towards increased proportions of gametophytes in the sheltered site clearings, and increased proportions of tetrasporophytes in the exposed site clearings. 2) More gametophytes were found than tetrasporophytes among the sheltered site recruits for all clearing times (Figure 3.4) suggesting that the stochastic model may be an appropriate tool for predicting phase proportions in that site. In the exposed site (Figure 3.5) for two sampling times more gametophytes than tetrasporophytes were found among the recruits, in the third time the phase proportions were similar to one another. At no time were the phase proportions among the recruits similar to those in the surrounding population suggesting that the stochastic model does not have sufficient predictive ability for the exposed site. 3) In the exposed site more tetrasporophytes recruited than the proportion of reproductive cystocarpic blades in the surrounding population at each clearing time, contrary to the Nobleweed-Pauperweed model; in the sheltered site more gametophytes recruited than the proportion of reproductive tetrasporophytes in the surrounding population at each clearing time, as predicted by the N-P model. By a loose definition of specialization it appears that tetrasporophytes were specialized for recruitment in the exposed site and gametophytes specialized for recruitment in the sheltered site. 4) Following recruitment, while there were not significant changes in the proportions of gametophytes and tetrasporophytes over time, there were apparent trends. In the sheltered site the proportion of gametophytes appeared to increase, contrary to the N-P model; in the exposed site the opposite appeared to be true, as predicted by the N-P model. In neither the exposed site nor the sheltered site was the Nobleweed-Pauperweed model supported, though for different reasons. 5) There was a significantly higher proportion of tetrasporophytes in the exposed site than in the sheltered site at all times of year.

While it is apparent that none of the models were completely successful in both sites at all times the tests of the models have given some direction for future research into the mechanisms responsible for the distributions of the gametophytes and tetrasporophytes of Mazzaella lilacina with wave exposure. It has long been recognized that the period of greatest mortality is between the release of spores and the appearance of macro-recruits (Chapman 1986). In the exposed site not as many gametophytes recruited as was expected based on the number of reproductive tetrasporophytes and their spore production (Chapter 1). In the sheltered site not as many tetrasporophytes recruited as was expected based on the number of reproductive cystocarpic blades. Obviously important processes were at work between spore release and their growth to .5 cm long blades. The pertinent factors may include herbivory by micro-herbivores, dessication, light tolerance, attachment strength, nutrient availability, and competition between the phases and with other taxa (Vadas et al. 1992).

A laboratory experiment to test the efficacy of micro-herbivores could involve the seeding of settling plates with a known density of carpospores and tetraspores followed by cultivation in the presence or absence of various herbivores.

# CHAPTER 4: RESISTANCE TO REMOVAL

## Introduction

If tetrasporophytes of Mazzaella lilacina (Postels and Ruprecht) Leister are more resistant to removal by wave action than gametophytes then populations should have an increasing proportion of tetrasporophytes as wave exposure increases. Dyck et al. (1985) observed that the proportion of tetrasporophytes in populations of M. lilacina (as Iridaea cordata), in Barkley Sound, British Columbia, increased with wave exposure. At the same time they observed that the proportion of gametophytes found in the drift was much higher than that found in the attached population, suggesting that the gametophytes were more susceptible to wave induced mortality.

Resistance to removal may be the result of 1) a morphology that is subject to less drag, 2) greater material strength of the stipe or holdfast, 3) a flexible thallus that may be extendible and therefore resistant to short bursts of force as well as able to press against the substrate and effectively decrease its surface area (Koehl 1986), or 4) thicker stipes. The extensibility of each phase's thalli was not dealt with directly by this research. Work has been done comparing the free living phases of M. lilacina (Shaughnessy and DeWreede Pers. Comm.). Dudgeon and Johnson (1992) made a biomechanical comparison of Chondrus crispus Stackhouse and Mastocarpus stellatus (Stack. in With.) Guiry, in the field, to determine which one was more susceptible to wave induced mortality. They found that in the mixed stands where these algae often occur C. crispus experienced more frequent dislodgement than M.

stellatus. Both species required the same mean force per unit of stipe cross sectional area to break, but C. crispus had sufficiently greater blade surface area so its mortality rate was significantly higher.

The primary directional force that an algal blade experiences when a wave hits is the result of form drag (Koehl 1986). As water flows around an object turbulence is created on the downstream side of that object, and the result is a force in the direction of flow, i.e. drag, which is roughly proportional to the square of the water velocity. Surge channels and subtidal habitats often have water velocities of 1 m/s, whereas intertidal habitats experience water velocities as high as 10 or 20 m/s during storms (Denny 1988). If the water velocity is high enough the drag may be sufficient to cause mechanical failure of algal thalli. Drag is also dependent on the shape, size, and rigidity of the object. Flexibility is often a detriment in the presence of a flowing fluid. Because of the turbulent eddies created by its moving surface a flag experiences much higher drag than does a weather vane of the same size and shape (Hoerner 1965 [fr. Vogel 1989]).

Studies by Carrington (1990) showed that the most important morphological character of macroscopic algae determining drag was planform area, which is the surface area of a two dimensional image of a blade. Shaughnessy (Pers. Comm.) found that in Mazzaella lilacina, from Barkley Sound, British Columbia, gametophytes were significantly wider than tetrasporic blades of the same length, especially in moderate to large sized blades.

The haploid and diploid phases of Mazzaella lilacina are known to have different carrageenans which may result in a difference in the material strengths of the two phases (McCandless et al. 1983, Waaland

1973). Strength is defined as the force required to cause a material to fail divided by the area of the point of failure (Denny 1988). A synonymous term for strength and the one used more often in this study is 'Stress to Break' ($S_b$). Tetrasporic thalli contain lambda carrageenan and gametophytes contain kappa carrageenan. Based solely on the laboratory properties of the two carrageenans, the kappa carrageenan containing gametophytes would be expected to be stronger than the lambda carrageenan containing tetrasporophytes. Gels from extracted kappa carrageenan are less viscous than those from lambda carrageenan. Waaland (1973) found that 61% of the dry weight of Iridaea cordata (now Mazzaella lilacina) was carrageenan, but it is not known whether it plays an important biomechanical role in strength determination.

The force required to break each blade and the cross sectional area of the breakage point were measured in situ and used to obtain the stress to break ($S_b$).

Applied stress ($S_a$) is an estimation of the drag that a particular thallus would experience divided by the cross sectional area of the stipe-holdfast junction. If the applied stress is greater than the stress to break ($S_b$) then the blade will be separated from the holdfast, most likely by failure of the stipe-holdfast junction (Carrington 1990).

The recommended equation (Carrington 1990) for drag is as follows;

$$\text{Drag} = 1/2\ C_d\ A\ p\ U^2$$

where $C_d$ is the drag coefficient, p the density of water, A the planform area, and U the velocity of water measured in meters per second.

The drag coefficient, $C_d$, is directly proportional to the water velocity raised to some exponent, usually referred to as E.

$C_d$ is proportional to $U^E$

For a bluff body, i.e. unstreamlined, at moderate to high water velocities the drag on the object is directly proportional to the square of water velocity. In other words the drag coefficient is constant with respect to water velocity so E is zero. If drag is independent of water velocity then E will be -2 because the drag coefficient must decrease at the same rate that $U^E$ increases if the drag is to remain constant. Many objects change shape in flowing water thereby effectively decreasing the amount of surface area exposed to the water so drag increases at something less than the square of water velocity. Objects that reconfigure will not have a constant drag coefficient, and the more an object reconfigures the more negative will be the E value. The E value can be used as an estimate of the extent of reconfiguration of an object (Vogel 1989, Carrington 1990).

Carrington (1990) calculated the drag coefficient for _Mazzaella flaccida_ (as _Iridaea_ _flaccida_) to be between 0.04 and 0.07 at a water velocity of 2.5 m/s. The E value was calculated as -0.76 with a standard deviation of 0.15.

The null hypotheses to be tested in this chapter are:

1) Tetrasporophytes are as likely to be removed by a wave of a particular velocity as gametophytes.

2) The material strength, measured as stress to break, is equal for tetrasporophytes and gametophytes.

2a) The average force to break tetrasporophytes is the same as the average force to break gametophytes.

2b) The average stipe-holdfast junction area of the tetrasporophytes is equal to that of the gametophytes.

3) The average blade area for tetrasporophytes is the same as that for gametophytes.

## Materials and Methods

For all of the questions addressed in this study sampling was done in April and June of 1993 at Second Beach in Barkley Sound, British Columbia. A transect line with randomly spaced marks was haphazardly placed in the intertidal *Mazzaella* zone. One blade from the holdfast nearest to each mark was placed in a clamp and pulled until it broke. The force to break was determined by attaching the clamp across the entire width of each blade near the end of the apophasis. A machine with a small electric motor and a spring scale was attached to the clamp. The electric motor applied a tension force to the thallus until it snapped. The maximum extension of the spring scale was recorded in Kilograms then converted to force, in Newtons, by multiplying it by acceleration due to gravity (9.8 $m/s^2$). In all cases breakage occurred within one second.

After removal the blades were air dried and taken to the laboratory. Following rehydration the planform area and stipe-holfast junction diameter were measured. Thalli were placed in sea water for between 10 and 15 minutes in order to completely rehydrate the stipe. Optimum rehydration times, i.e. the time required to approach as closely as possible the original dimensions, were based on work by Shaughnessy (Pers. Comm.) for *Mazzaella lilacina*. He has calculated optimum rehydration times by comparing freshly picked blades and the same blades after they had been dried and rehydrated for various lengths of time. Blades from the April sampling time were measured and photocopied, for use in finding their planform areas, before being dehydrated for transport.

In approximately 80% of the samples taken the break occurred at the junction between the stipe and holdfast. The junction in *Mazzaella lilacina* is clearly visible as a short light-green segment at the end of the stipe. The other 20% of structural failures occurred in either the stipe or blade, or the holdfast was removed from the substratum A small percentage of individuals were removed intact with a portion of the underlying rock still attached to the holdfast. Because the vast majority of blades separate from the holdfast at the stipe-holdfast junction and because of the difficulties in standardizing measurements from different parts of the blade or stipe the comparison of material strengths used only those individuals which failed at the stipe-holdfast junction.

To determine the planform area, to be used in calculating drag, each blade was pressed between two sheets of glass and photocopied. The copied areas were then cut out, weighed, and the weights compared to the weight of a known area of paper.

The junction between the stipe and holdfast is roughly circular in cross section so its area, in square millimeters, was determined by measuring the diameter of the point of failure and using this measurement in the equation for the area of a circle. In those cases where the stipe cross-section was clearly an ellipse the recorded diameter was intermediate between the long and short dimensions.

In April random samples were taken from both populations (Sheltered and Exposed). In June, however, due to the limited time available samples were only taken from the exposed site. Because the vegetative phases are indistinguishable in the field and are present in varying proportions there are varying numbers of observations per phase both within and between sites. Phase was determined after the field experiments by testing a portion of each thallus using the Acetal-Resorcinol colorimetric test (Garbary and DeWreede 1988).

Each hypothesis was tested using blades of all sizes together and then using only blades longer than 15cm The rationale for this was that it was only in the larger blades that a significant difference in the width of the phases occurred (Shaughnessy Pers. Comm.).

Hypothesis 1, that tetrasporophytes are as likely as gametophytes to be removed by a wave of a particular velocity, was tested by plotting water velocity against the probability of a blade being dislodged. Confidence intervals were placed around the curves for gametophytes and tetrasporophytes. The route to this final presentation of the data required three steps, which are outlined below. A great deal of appreciation is due to Mark Denny for guidance in the presentation of the resistance to removal data (Figures 4.1-4.12).

The first step required that the forces required to break ($F_b$), as measured in the field experiments, be ordered from smallest $F_b$ to largest. Each $F_b$ was then ranked by dividing case by n+1, where case refers to each observation in the series from 1 to n, the last observation. The ranks so produced were uncorrected probabilities of dislodgement, and these data were plotted against their corresponding forces to break producing a sinusoidal curve.

Using the nonlin module of SYSTAT (Wilkinson 1990) a curve was fitted to the data according to the equation;

$$\text{Estimate} = \exp ( -1 * (( a-b*F_b )/( a-b*e ))\char`^( 1/b )).$$

Where Estimate is the best estimate of each probability of dislodgement based on the fitted curve. $F_b$ is the force to break measured in the field, while a, b, and e are parameters created to make the curve fit, by the computer, and refer to alpha, beta and epsilon respectively.

A second curve was created by plotting the average drag experienced by the population against water velocity. As previously mentioned drag is partially dependent on planform area. Both the gametophytes and tetrasporophytes had upper and lower confidence limits based on the 95% confidence limits for planform area.

The final curve was a combination of the previous two curves. Instead of using Fb in the first equation drag was used. So, for each water velocity a probability of dislodgement was produced, with upper and a lower 95% confidence limits for each phase

All of the non-graphical data were analyzed using simple linear regressions. The experiment was set up as a completely randomized design with unequal numbers of replicates in each factor, the sheltered and exposed sites being the two factors.

Since the design was unbalanced because of the unequal numbers of observations in each factor, dummy variables were used.

In order to meet the regression assumptions of normally distributed data and homogeneity of variance some of the data were log transformed.

## Results

### Hypothesis 1: Removal of tet. <> removal of gam.

### Whole Population

The progression of graphs leading to and including the Probability of Removal vs. Water Velocity graph are presented (Figures 4.1-4.12). For the sheltered site in April and the exposed site in June, sets of graphs were constructed using both the whole sampled population and also only blades longer than 15 cm.

Figure 4.1: For the sheltered site, the proportion of gametophytes and tetrasporophytes which would be broken with a given force in extension. Force to break (MegaNewtons) was calculated from field measurements made in April 1993.

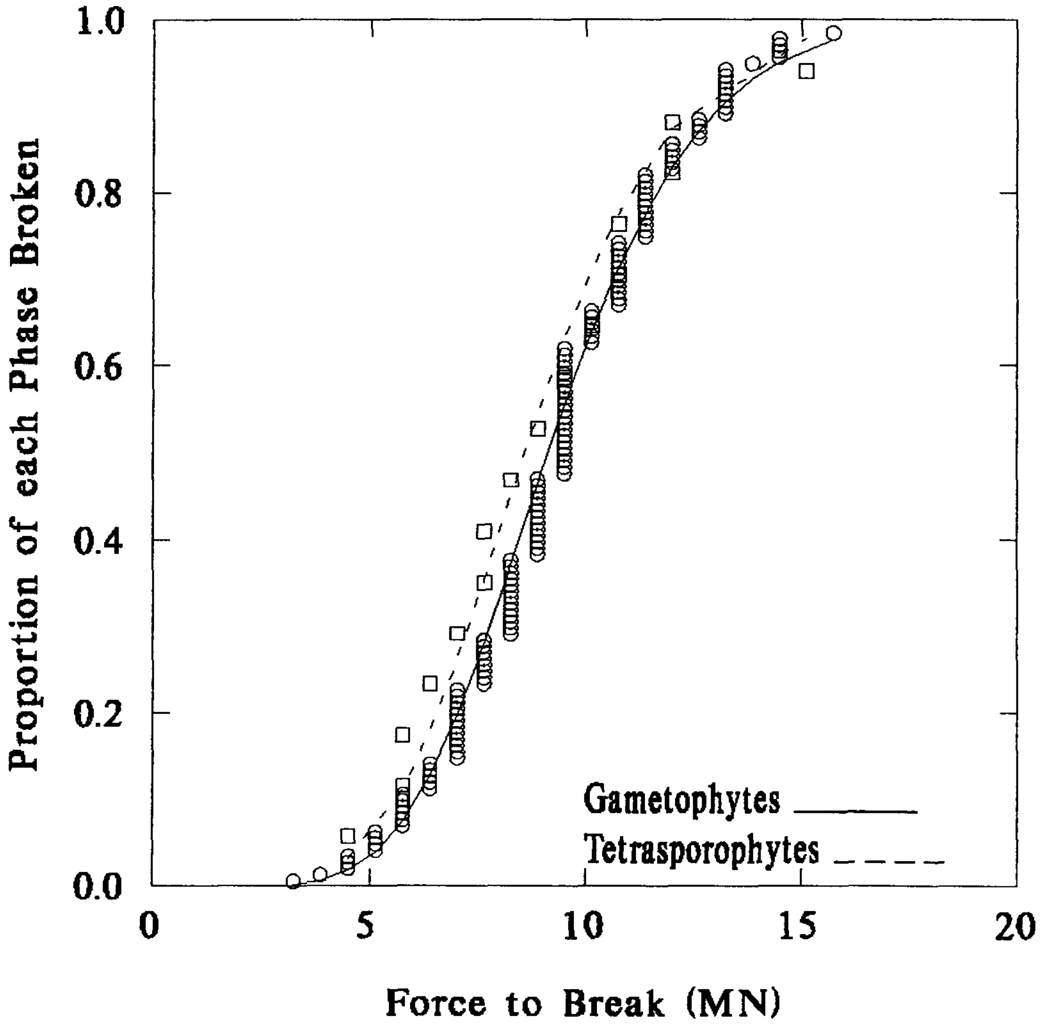


Figure 4.2: For the sheltered site, the drag experienced by gametophytes and tetrasporophytes with a given water velocity. The upper and lower 95% confidence limits for blade area were used in calculating drag. Measurements were taken in April 1993.

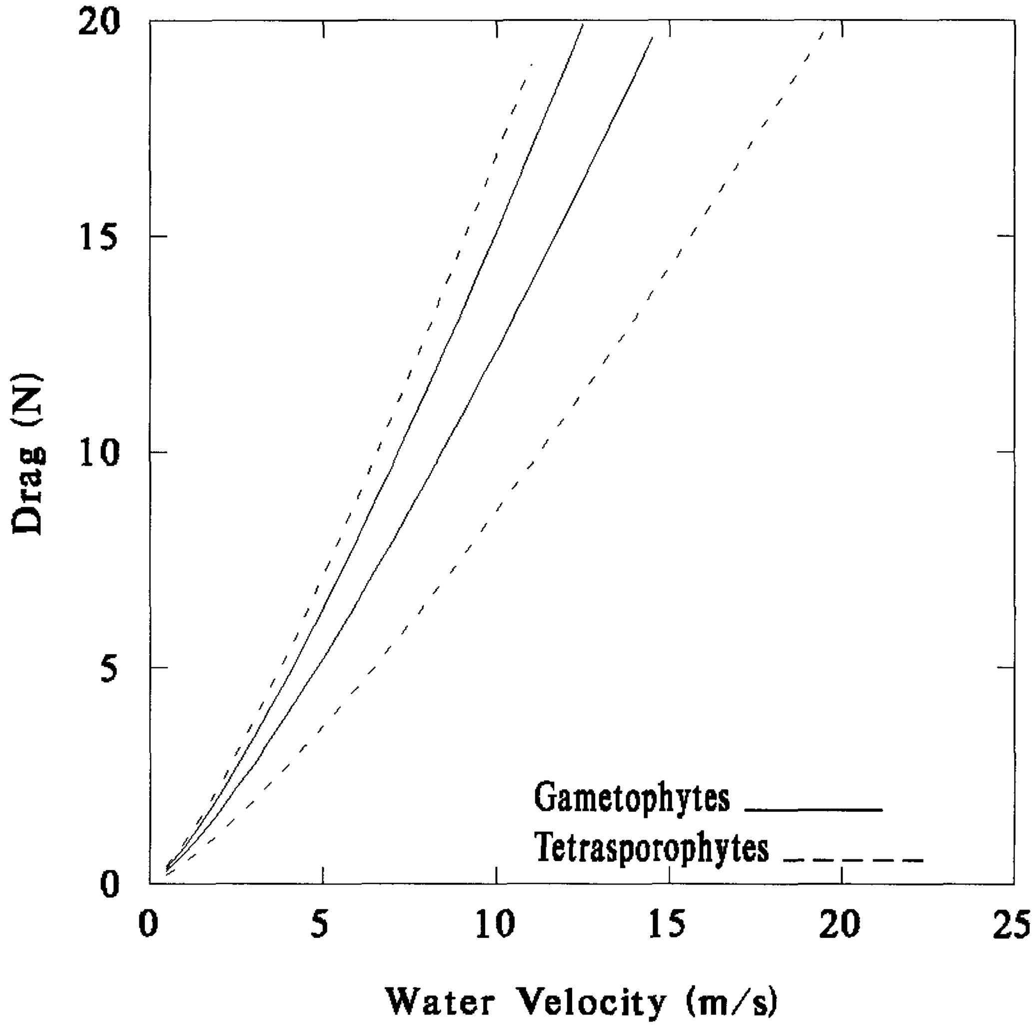


Figure 4.3: For the sheltered site, the proportion of the gametophytes and tetrasporophytes which would be removed from the substrate with water of a given velocity. Blade measurements were taken in April 1993.

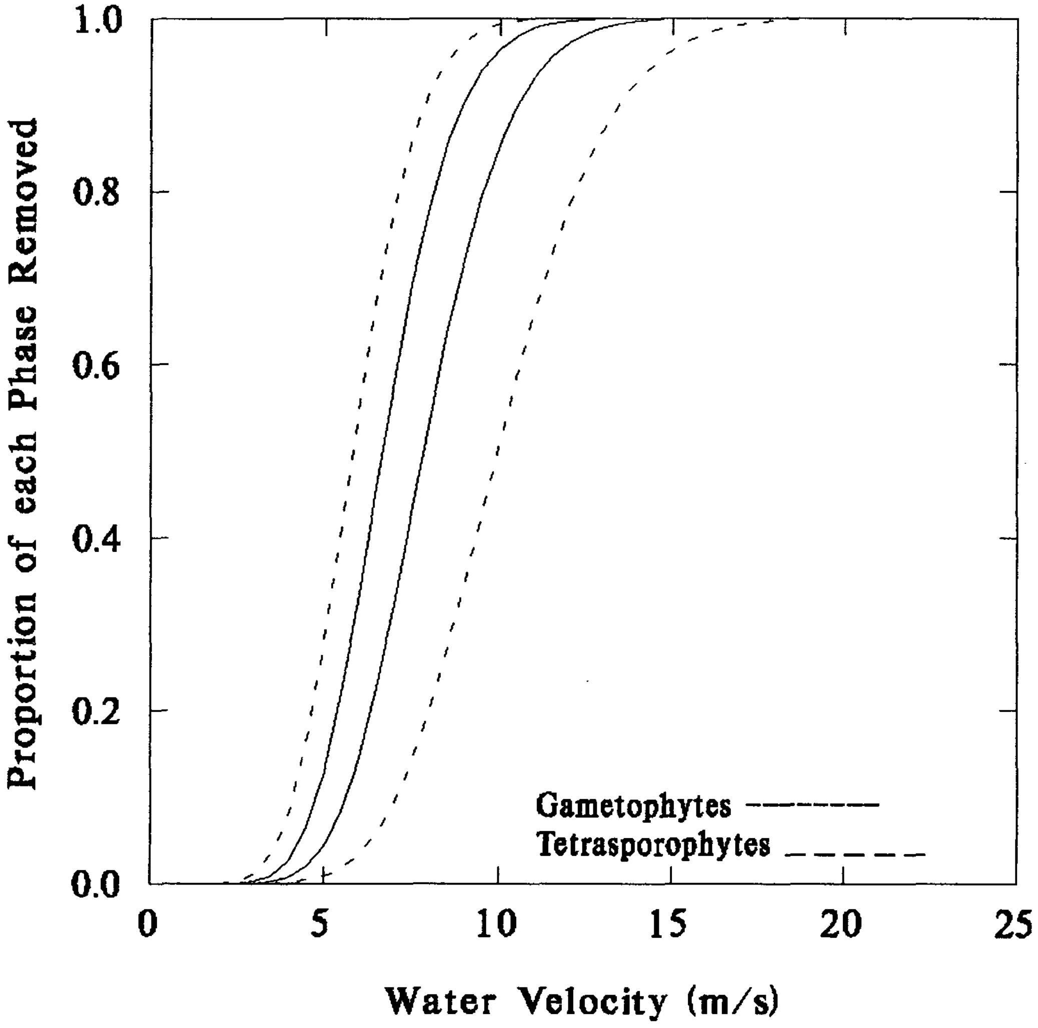


Figure 4.4: For the exposed site, the proportion of gametophytes and tetrasporophytes which would be broken with a given force in extension. Force to break (MegaNewtons) was calculated from field measurements made in June 1993.

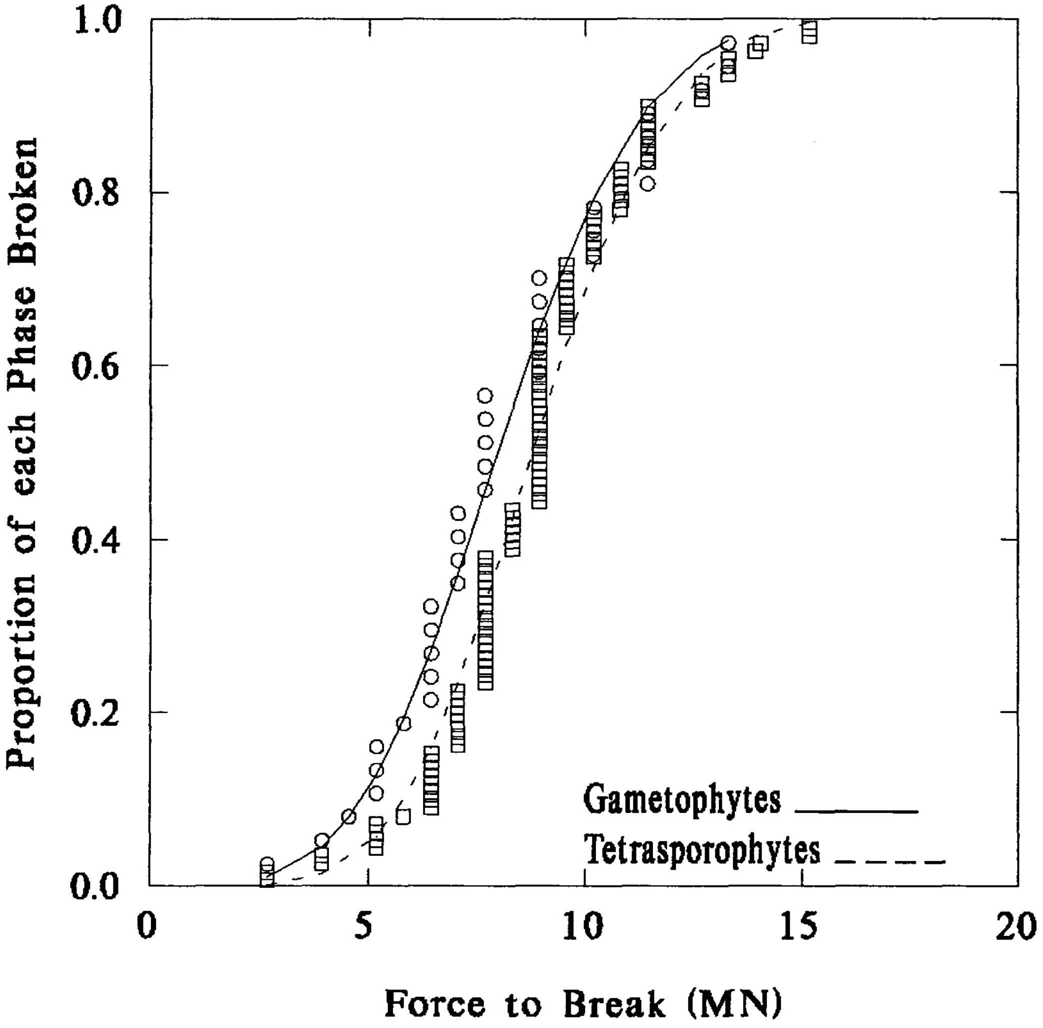


Figure 4.5: For the exposed site, the drag experienced by gametophytes and tetrasporophytes with a given water velocity. The upper and lower 95% confidence limits for blade area were used in calculating drag. Measurements were taken in June 1993.

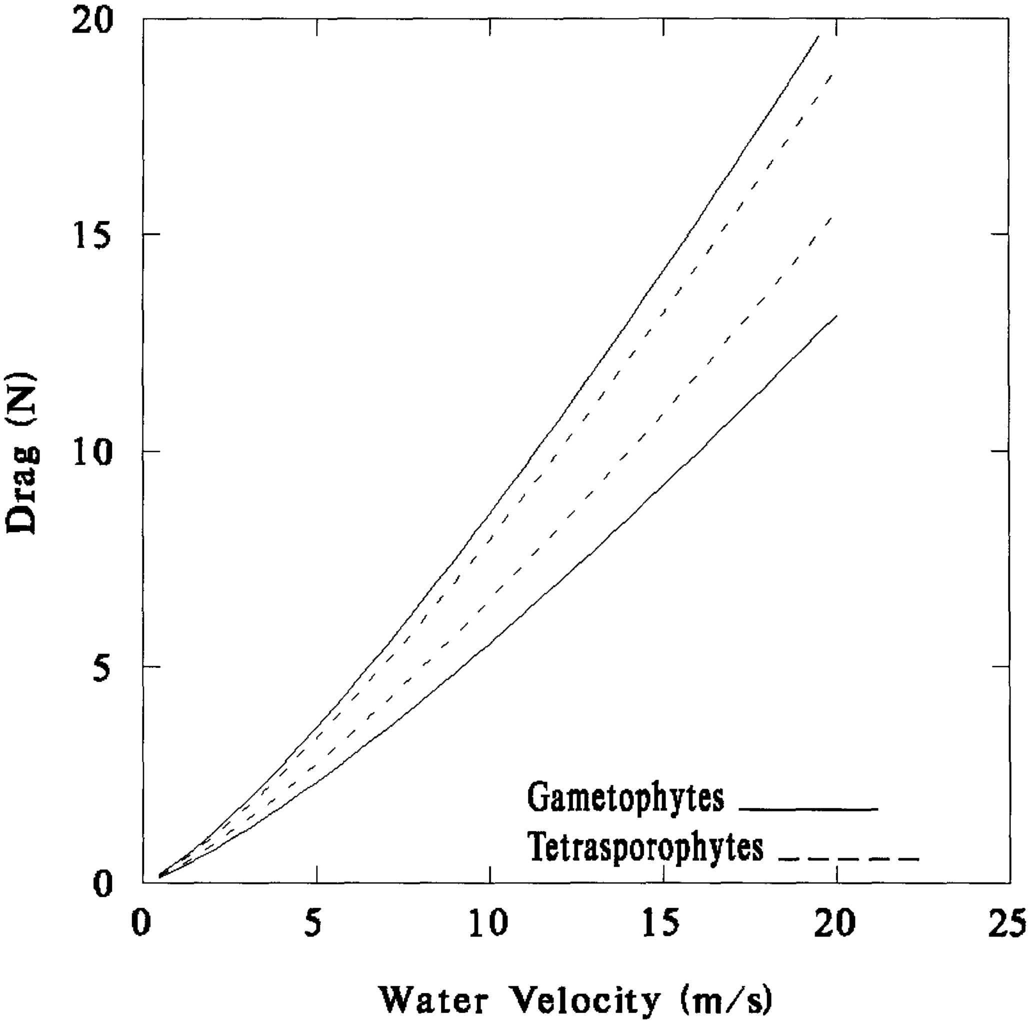


Figure 4.6: For the exposed site, the proportion of the gametophytes and tetrasporophytes which would be removed from the substrate with water of a given velocity. Blade measurements were taken in June 1993.

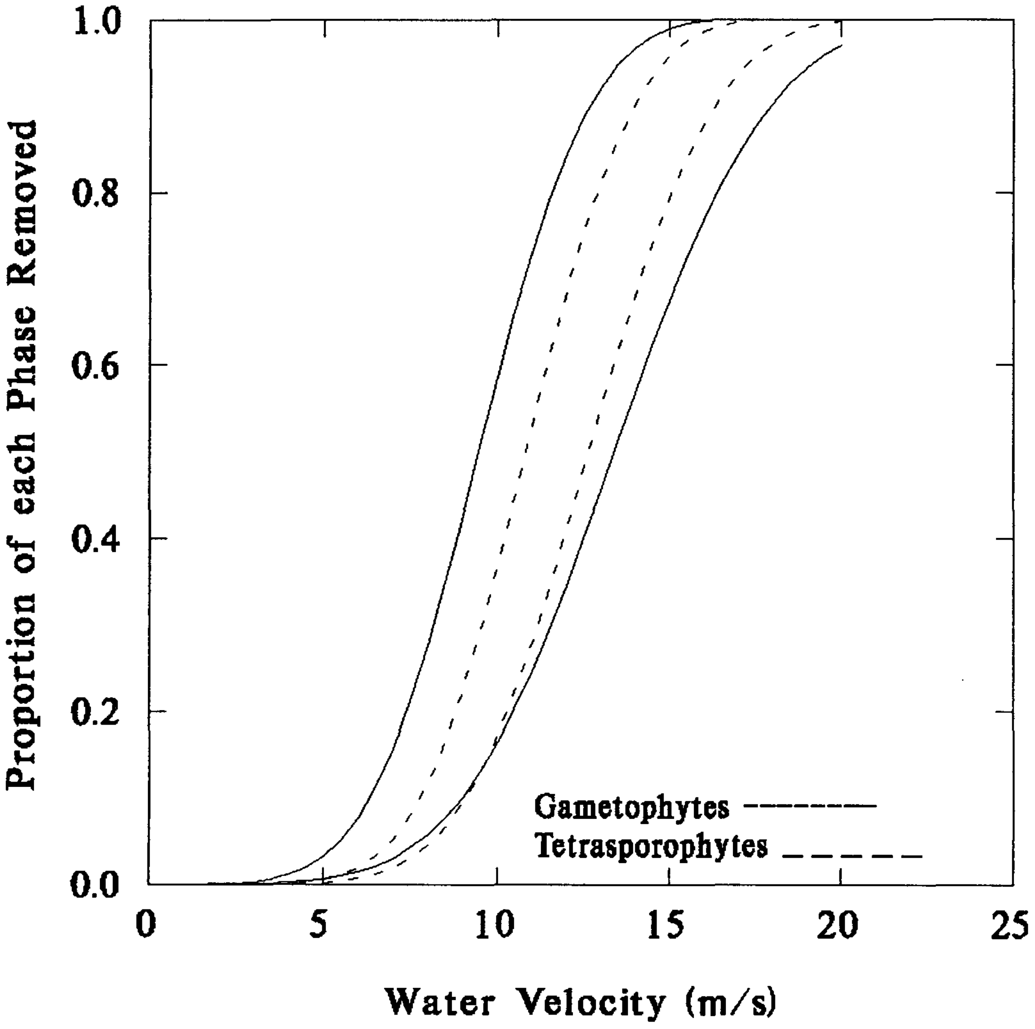


Figure 4.7: For the sheltered site, the proportion of gametophytes and tetrasporophytes longer than 15cm which would be broken with a given force in extension. Force to break (MegaNewtons) was calculated from field measurements made in April 1993.

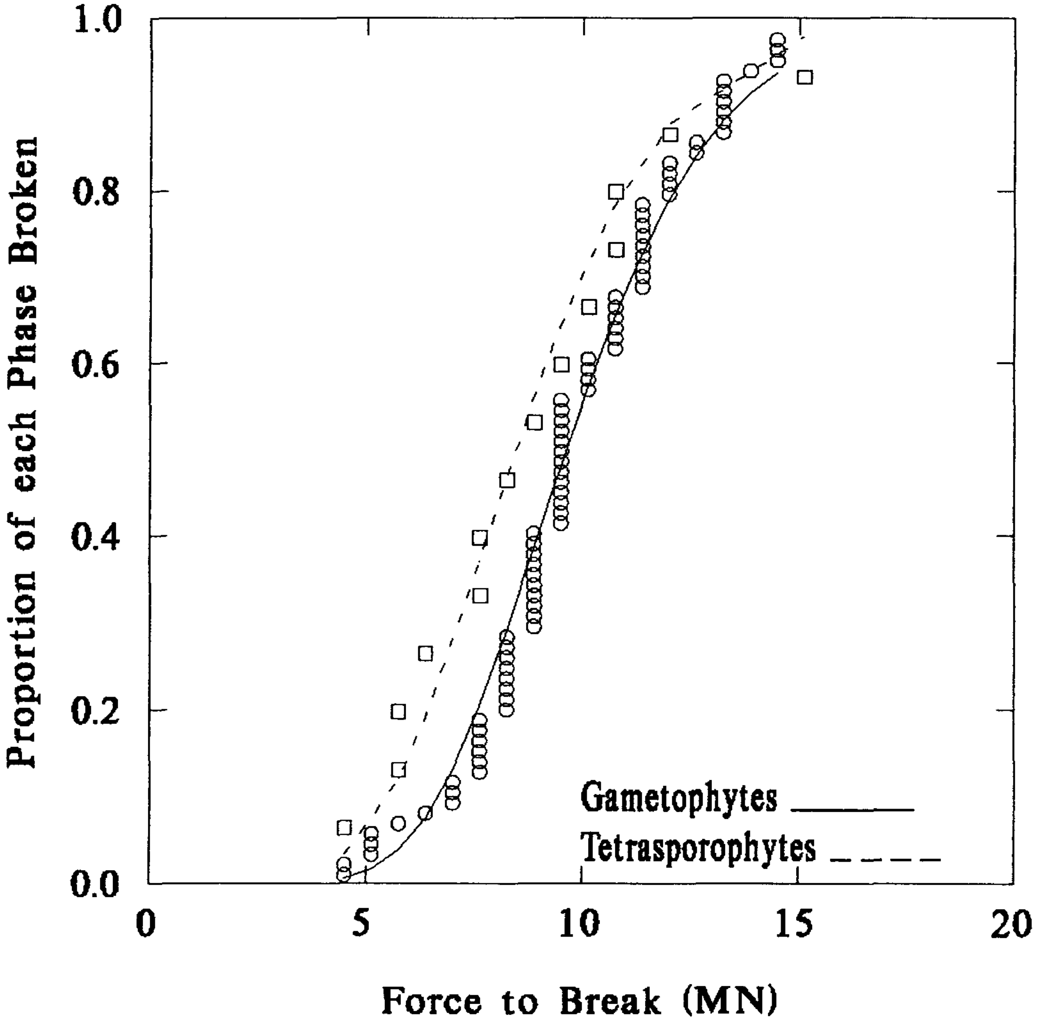


Figure 4.8: For the sheltered site, the drag experienced by gametophytes and tetrasporophytes longer than 15cm with a given water velocity. The upper and lower 95% confidence limits for blade area were used in calculating drag. Measurements were taken in April 1993.

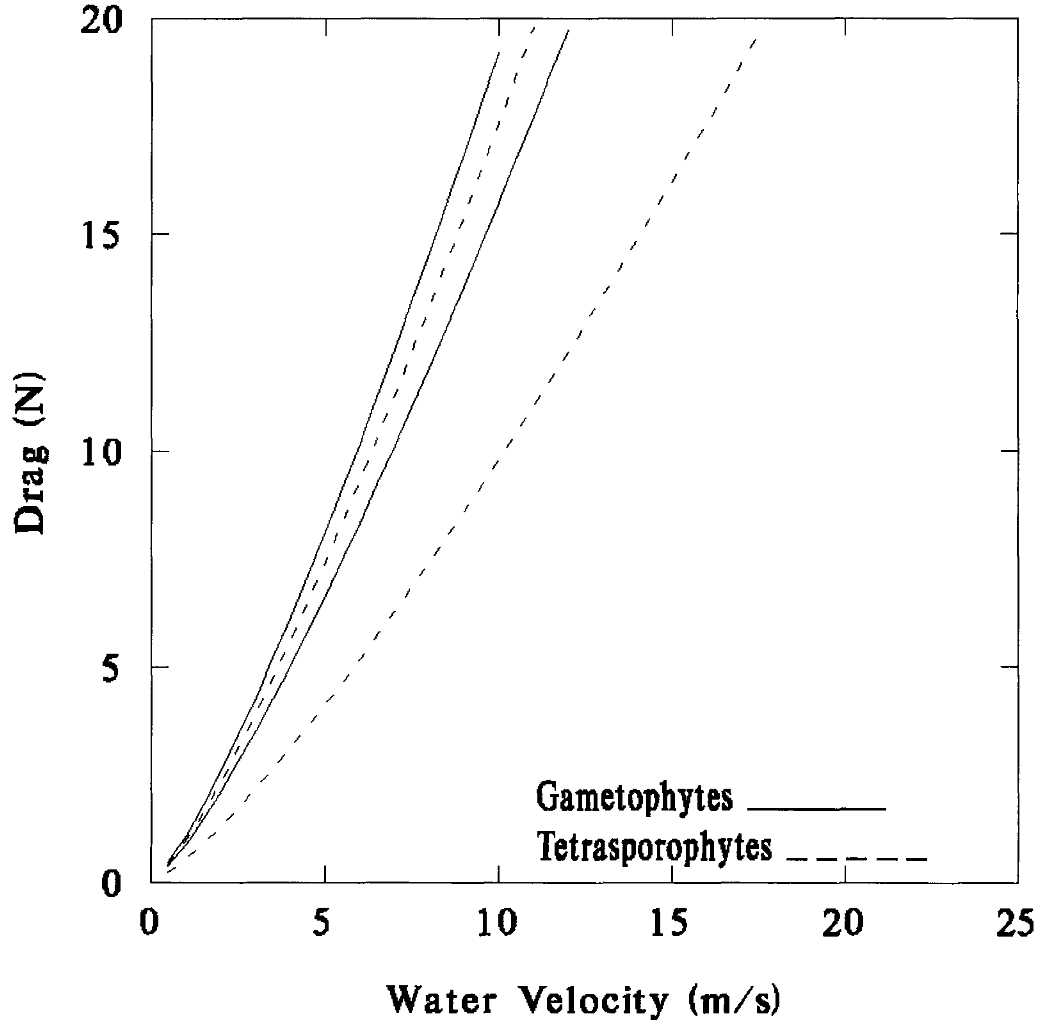


Figure 4.9: For the sheltered site, the proportion of the gametophytes and tetrasporophytes longer than 15cm which would be removed from the substrate with water of a given velocity. Blade measurements were taken in April 1993.

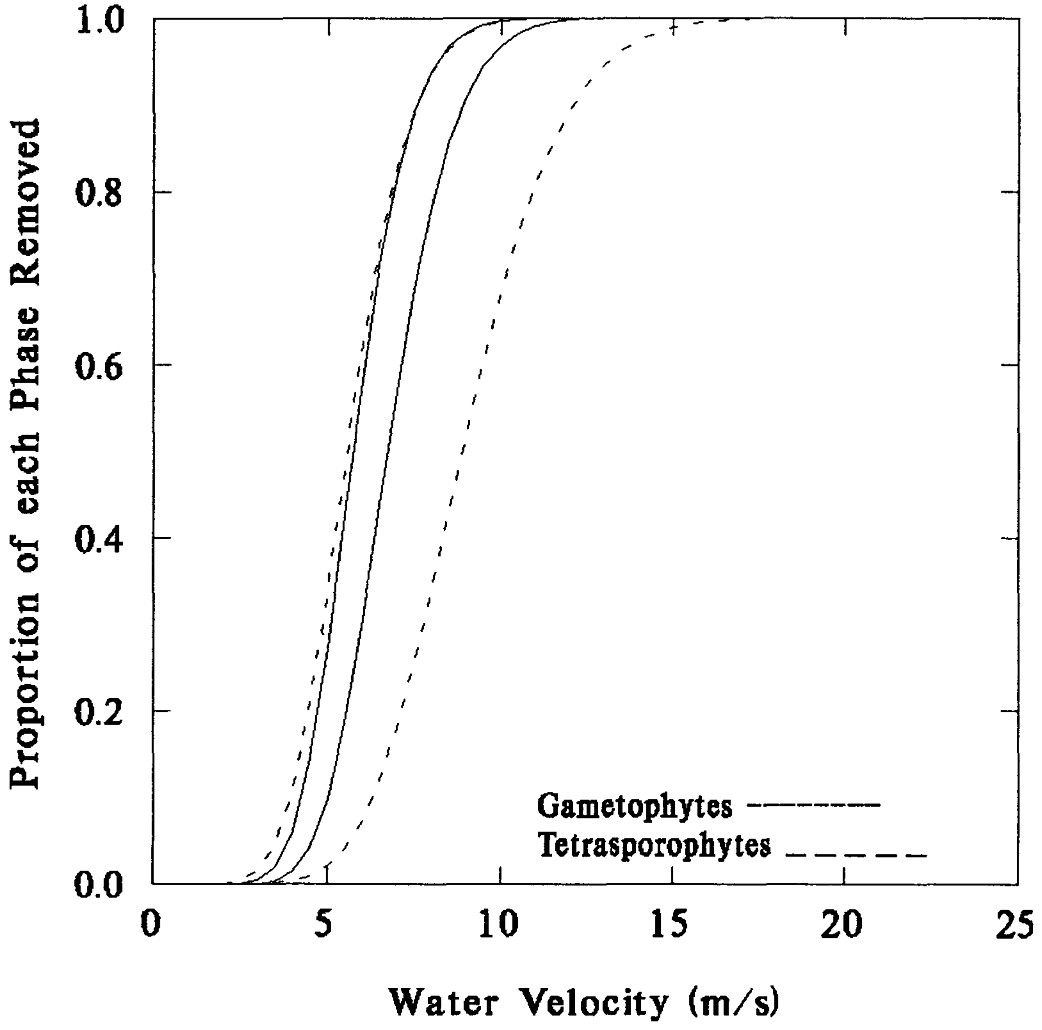


Figure 4.10: For the exposed site, the proportion of gametophytes and tetrasporophytes longer than 15cm which would be broken with a given force in extension. Force to break (MegaNewtons) was calculated from field measurements made in June 1993.

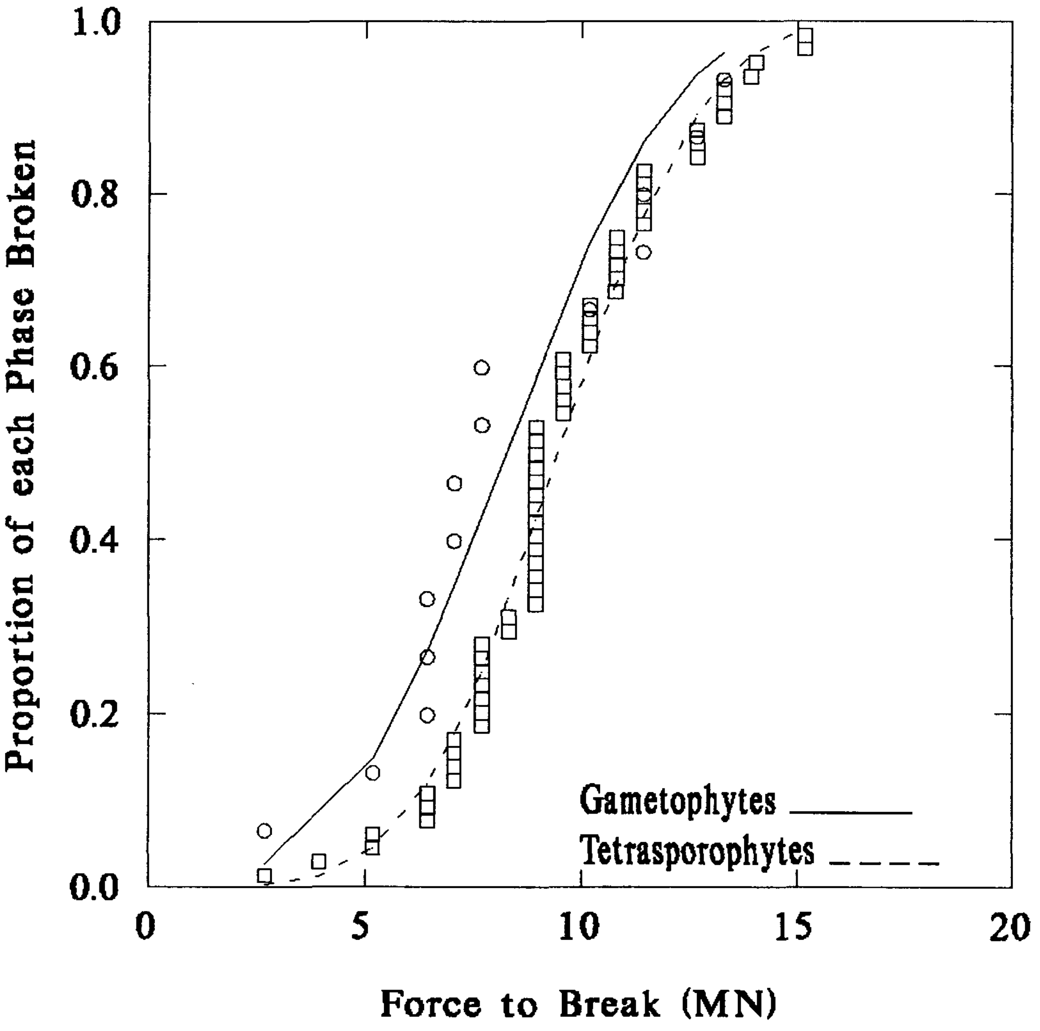


Figure 4.11: For the exposed site, the drag experienced by gametophytes and tetrasporophytes longer than 15cm with a given water velocity. The upper and lower 95% confidence limits for blade area were used in calculating drag. Measurements were taken in June 1993.

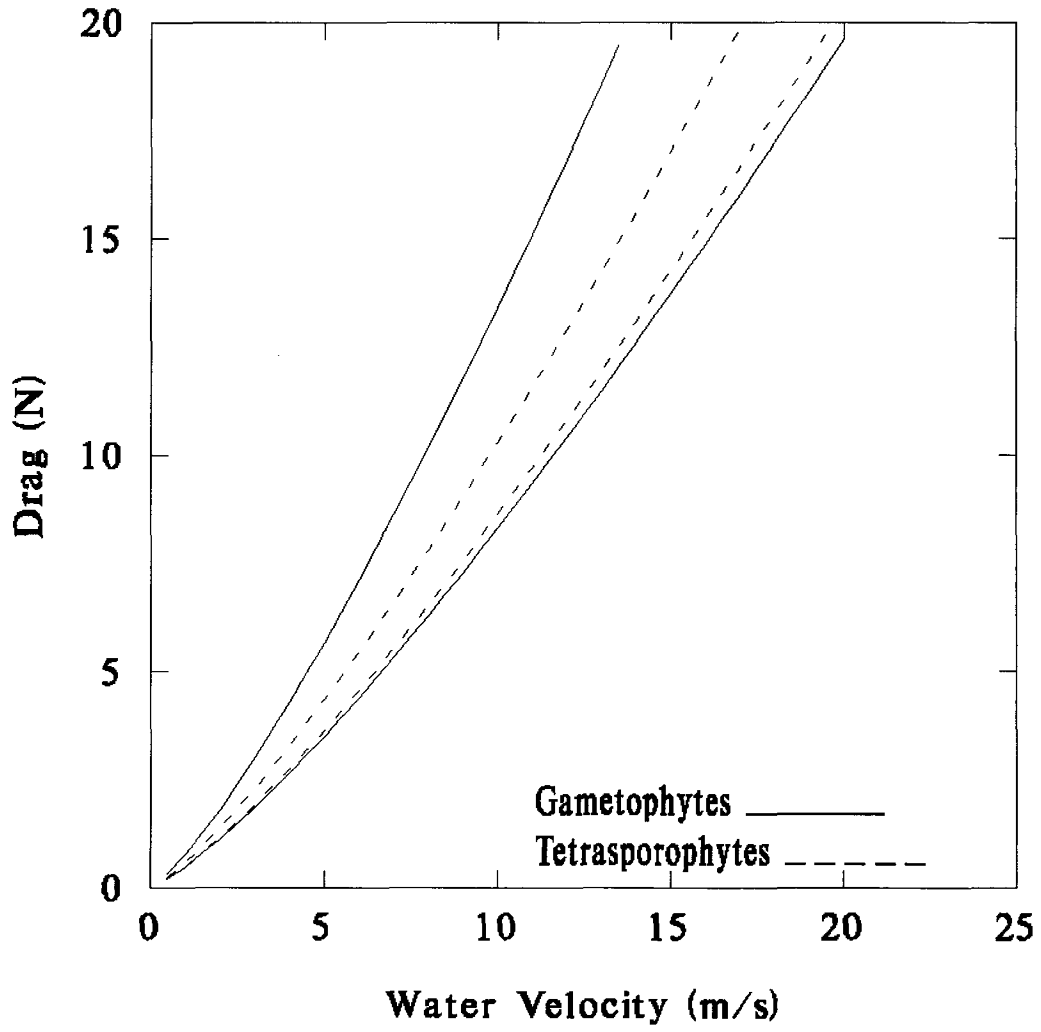
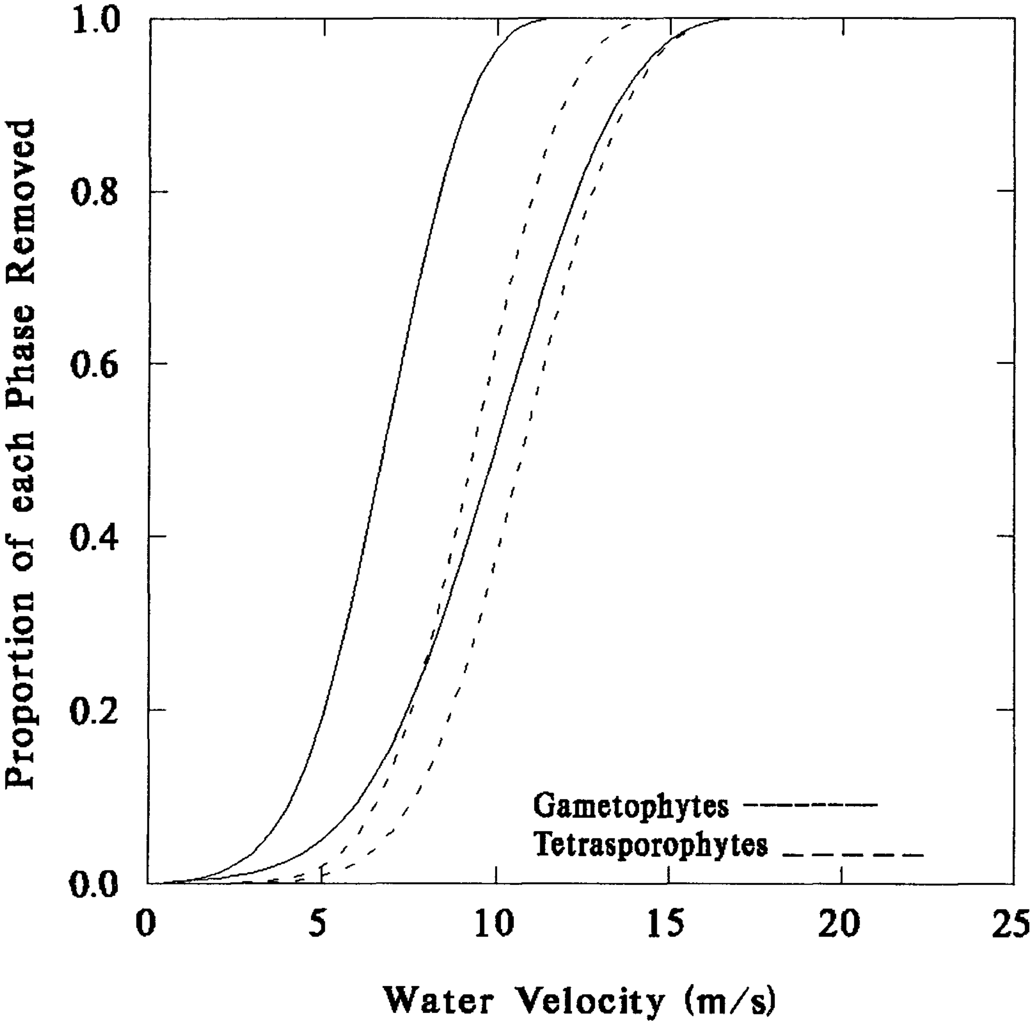


Figure 4.12: For the exposed site, the proportion of the gametophytes and tetrasporophytes longer than 15cm which would be removed from the substrate with water of a given velocity. Blade measurements were taken in June 1993.



Hypothesis 2: $S_b$ of tet. <> $S_b$ of gam.

Whole Population

The regression for stress to break ($S_b$) on phase for the sheltered site at the April sampling time had an $R^2$ value of 0.001. The data were log transformed.

ANALYSIS OF VARIANCE

| SOURCE | SUM-OF SQUARES | DF | MS | F-RATIO | P |
|---|---|---|---|---|---|
| PHASE | 0.058 | 1 | 0.058 | 0.101 | 0.751 |
| ERROR | 59.267 | 104 | 0.570 | | |

The mean values for the gametophytes and tetrasporophytes were 1.85 $MN/mm^2$ and 1.78 $MN/mm^2$, respectively. The null hypothesis of no difference was accepted. An Alpha value of 0.05 was used

At the June sampling time the regression on $S_b$ for the exposed site had an $R^2$ value of 0.003.

ANALYSIS OF VARIANCE

| SOURCE | SUM-OF-SQUARES | DF | MS | F-RATIO | P |
|---|---|---|---|---|---|
| PHASE | 5.419 | 1 | 5.419 | 0.324 | 0.570 |
| ERROR | 1655.062 | 99 | 16.718 | | |

The null hypothesis of no difference between the gametophytes and tetrasporophytes was accepted. The phases had mean Stresses to Break of 7.56 MN/mm$^2$ and 6.99 MN/mm$^2$, respectively.

### Large Blades Only

At the April sampling time the regression on $S_b$ for the sheltered site had an $R^2$ value of 0.001. The data was log transformed.

ANALYSIS OF VARIANCE

| SOURCE | SUM-OF-SQUARES | DF | MS | F-RATIO | P |
|---|---|---|---|---|---|
| PHASE | 0.021 | 1 | 0.021 | 0.033 | 0.856 |
| ERROR | 42.292 | 99 | 0.641 | | |

The null hypothesis of no difference between the gametophytes and tetrasporophytes was accepted. The phases had mean Stresses to Break of 1.84 MN/mm$^2$ and 1.79 MN/mm$^2$, respectively.

At the June sampling time the regression on $S_b$ for the sheltered site had an $R^2$ value of 0.025.

ANALYSIS OF VARIANCE

| SOURCE | SUM-OF-SQUARES | DF | MS | F-RATIO | P |
|---|---|---|---|---|---|
| PHASE | 25.060 | 1 | 25.060 | 1.369 | 0.247 |
| ERROR | 988.499 | 99 | 18.306 | | |

The null hypothesis of no difference between the gametophytes and tetrasporophytes was accepted. The phases had mean Stresses to Break of 8.90 MN/mm$^2$ and 6.87 MN/mm$^2$, respectively.

Hypothesis 2a: The $F_b$ for tet. <> the $F_b$ for gam.

Whole Population

The regression of force to break (Fb) on phase for the sheltered site in April had an $R^2$ value of 0.003.

ANALYSIS OF VARIANCE

| SOURCE | SUM-OF-SQUARES | DF | MS | F-RATIO | P |
|---|---|---|---|---|---|
| PHASE | 3.749 | 1 | 3.749 | 0.498 | 0.481 |
| ERROR | 1150.867 | 153 | 7.522 | | |

The mean forces to break for the gametophytes and tetrasporophytes were 9.38 MN and 8.87 MN, respectively. The null hypothesis of no difference was accepted.

The regression for the exposed site in June had an $R^2$ value of 0.017.

ANALYSIS OF VARIANCE

| SOURCE | SUM-OF-SQUARES | DF | MS | F-RATIO | P |
|---|---|---|---|---|---|
| PHASE | 15.068 | 1 | 15.068 | 2.523 | 0.114 |
| ERROR | 853.867 | 143 | 5.971 | | |

The null hypothesis of no difference between the means was accepted. The mean values for the force to break for gametophytes and tetrasporophytes were 8.10 MN and 8.85 MN, respectively.

### Large Blades Only

The regression of force to break on phase for the sheltered site in April had an $R^2$ value of 0.020. The data were log transformed.

ANALYSIS OF VARIANCE

| SOURCE | SUM-OF-SQUARES | DF | MS | F-RATIO | P |
|---|---|---|---|---|---|
| PHASE | 14.569 | 1 | 14.569 | 1.980 | 0.163 |
| ERROR | 698.925 | 95 | 7.357 | | |

The null hypothesis of no difference between the means was accepted. The mean values for the force to break for gametophytes and tetrasporophytes were 9.58 MN and 8.33 MN, respectively.

The regression for the exposed site in June had an $R^2$ value of 0.030.

ANALYSIS OF VARIANCE

| SOURCE | SUM-OF-SQUARES | DF | MS | F-RATIO | P |
|---|---|---|---|---|---|
| PHASE | 16.323 | 1 | 16.323 | 2.331 | 0.131 |
| ERROR | 525.251 | 75 | 7.003 | | |

The null hypothesis of no difference between the means was accepted. The mean values for the force to break for gametophytes and tetrasporophytes were 8.26 MN and 9.46 MN, respectively.

Hypothesis 2b: The s-h junction area for tet. <> that for gam.

Whole Population

The regression of stipe-holdfast junction area on phase for the sheltered site in April had an $R^2$ value less than 0.000. The data were log transformed.

ANALYSIS OF VARIANCE

| SOURCE | SUM-OF-SQUARES | DF | MS | F-RATIO | P |
|---|---|---|---|---|---|
| PHASE | 0.003 | 1 | 0.003 | 0.010 | 0.920 |
| ERROR | 34.941 | 103 | 0.339 | | |

The means for the gametophytes and tetrasporophytes were 0.34 $mm^2$ and 0.36 $mm^2$, respectively. The null hypothesis was accepted.

The regression for the area of the stipe-holdfast junction on phase for the exposed site in June had an $R^2$ value of 0.014. The data were log transformed.

ANALYSIS OF VARIANCE

| SOURCE | SUM-OF-SQUARES | DF | MS | F-RATIO | P |
|---|---|---|---|---|---|
| PHASE | 0.443 | 1 | 0.443 | 1.454 | 0.231 |
| ERROR | 30.171 | 99 | 0.305 | | |

The means for the gametophytes and tetrasporophytes were 0.38 $mm^2$ and 0.22 $mm^2$, respectively. The null hypothesis of no difference between the means was accepted.

### Large Blades Only

The regression of the area of the stipe-holdfast junction on phase for the sheltered site in April had an $R^2$ value of less than 0.000.

ANALYSIS OF VARIANCE

| SOURCE | SUM-OF-SQUARES | DF | MS | F-RATIO | P |
|---|---|---|---|---|---|
| PHASE | 0.001 | 1 | 0.001 | 0.001 | 0.976 |
| ERROR | 65.914 | 65 | 1.014 | | |

The means for the gametophytes and tetrasporophytes were 1.68 $mm^2$ and 1.69 $mm^2$, respectively. The null hypothesis of no difference between the means was accepted.

The regression for the exposed site in June had an $R^2$ value of 0.035. The data were log transformed.

ANALYSIS OF VARIANCE

| SOURCE | SUM-OF-SQUARES | DF | MS | F-RATIO | P |
|---|---|---|---|---|---|
| PHASE | 0.555 | 1 | 0.555 | 1.943 | 0.169 |
| ERROR | 15.416 | 54 | 0.285 | | |

The means for the gametophytes and tetrasporophytes were 0.14 $mm^2$ and .44 $mm^2$, respectively. The null hypothesis of no difference between the means was accepted.

<u>Hypothesis 3:</u> The blade area for tet. <> that for gam.

<u>Whole Population</u>

The regression for blade area on phase for the sheltered site had an $R^2$ value of 0.004.

ANALYSIS-OF-VARIANCE

| SOURCE | SUM-OF-SQUARES | DF | MS | F-RATIO | P |
|---|---|---|---|---|---|
| PHASE | 0.000 | 1 | 0.000 | 0.553 | 0.458 |
| ERROR | 0.010 | 153 | 0.000 | | |

The means for the gametophytes and tetrasporophytes were 0.014 $m^2$ and 0.013 $m^2$. The null hypothesis of no difference between the means was accepted.

The regression for the exposed site in June had an $R^2$ value of 0.001.

ANALYSIS-OF-VARIANCE

| SOURCE | SUM-OF-SQUARES | DF | MS | F-RATIO | P |
|---|---|---|---|---|---|
| PHASE | 0.000 | 1 | 0.000 | 0.076 | 0.783 |
| ERROR | 0.000 | 143 | 0.000 | | |

The means for the gametophytes and tetrasporophytes were 0.007 $m^2$ and 0.007 $m^2$. The null hypothesis of no difference between the means was accepted.

## Large Blades Only

The regression for blade area on phase for the sheltered site in April, has an $R^2$ value of 0.028.

ANALYSIS-OF-VARIANCE

| SOURCE | SUM-OF-SQUARES | DF | MS | F-RATIO | P |
|---|---|---|---|---|---|
| PHASE | 0.000 | 1 | 0.000 | 2.771 | 0.099 |
| ERROR | 0.006 | 95 | 0.000 | | |

The means for the gametophytes and tetrasporophytes were 0.018 $m^2$ and 0.014 $m^2$. The null hypothesis of no difference between the means was accepted, although the difference is approaching significance with the gametophytes having the larger mean blade area.

The regression for the exposed site in June had an $R^2$ value of 0.020. The data were log transformed.

ANALYSIS-OF-VARIANCE

| SOURCE | SUM-OF-SQUARES | DF | MS | F-RATIO | P |
|---|---|---|---|---|---|
| PHASE | 0.183 | 1 | 0.183 | 1.502 | 0.224 |
| ERROR | 9.127 | 75 | 0.122 | | |

The means for the gametophytes and tetrasporophytes were 0.009 $m^2$ and 0.010 $m^2$. The null hypothesis of no difference between the means was accepted.

### Comparison of Length and Width

#### Large Blades Only

Shaughnessy (Pers. Comm.) has asserted that, for thalli of the same length, and longer than 15cm, gametophytes are wider than tetrasporophytes. To test this assertion linear regressions of length on phase and width on phase were done. Thalli sampled in the sheltered site in April, 1993, were used.

Length on Phase

ANALYSIS-OF-VARIANCE

| SOURCE | SUM-OF-SQUARES | DF | MS | F-RATIO | P |
|---|---|---|---|---|---|
| PHASE | 6.661 | 1 | 6.661 | 0.528 | 0.469 |
| ERROR | 1085.159 | 86 | 12.618 | | |

The null hypothesis of no difference between the means was accepted. The mean lengths for the gametophytes and tetrasporophytes were 19.84 cm and 20.62 cm, respectively.

Width on Phase

ANALYSIS-OF-VARIANCE

| SOURCE | SUM-OF-SQUARES | DF | MS | F-RATIO | P |
|---|---|---|---|---|---|
| PHASE | 82.963 | 1 | 82.963 | 5.363 | 0.023 |
| ERROR | 1330.310 | 86 | 15.469 | | |

The null hypothesis of no difference between the means was rejected. The mean widths for the gametophytes and tetrasporophytes were 12.81 cm and 10.08 cm, respectively, with the gametophytes being wider.

## Discussion

A significant difference between the gametophytes and tetrasporophytes of Mazzaella lilacina in their probabilities of removal with a given water velocity would provide a mechanism by which the observed phase distributions with wave exposure could be explained. If the gametophytes are more likely to be removed than tetrasporophytes then a population of blades in a wave exposed area could become tetrasporophyte dominated. Given that approximately 10% of the thalli failed at the substratum, leaving none or only a small portion of the holdfast behind, the population of holdfasts as well as blades could become tetrasporophyte dominated.

Significant differences taken from field studies may be difficult to find because the blades which are measured are the ones that have survived to date and therefore represent those which are less likely to be removed by waves. By sampling during a time of year when storms are less common (i.e. late Spring and Summer, as done in this study) more of the thalli with low resistance to removal by waves may be present.

From the two 'Probability of Removal vs. Water Velocity' curves using the whole population (Fig. 3,6) it can be seen that there was complete overlap of the confidence bands for the gametophytes and tetrasporophytes. There were no significant differences in the proportions of gametophytes and tetrasporophytes removed at any water velocity. For the April sampling time in the sheltered site (Fig. 3) the confidence band for the tetrasporophytes completely encompassed the band for the gametophytes. One reason for the wider band for the tetrasporophytes at this sampling time was the lower number of samples used (16 vs. 139 for the gametophytes). In the case of the June sampling time in the exposed site (Fig. 6) it was the gametophytes which had the wider confidence band, completely enclosing the tetrasporophyte.

Thus, based on data from the whole population, there was no significant difference between the phases in the proportion of the population which would be removed with a given water velocity.

The two most influential ingredients that go into making the above curves are the mean planform areas of each phase, and the forces required to break the thalli, obtained in the field. Greater forces required to break the thalli will move the final curves to the right, as will smaller planform areas.

The shapes of the "Force to Break vs. Proportion of Population Removed" curves (Figs. 1,4,7,10) were responsible for the shapes of the final "Probability of Removal vs Water Velocity" curves (Figs. 3,6,9,12). For example, if very few tetrasporophytes were removed with low applied forces then the "Force to Break ..." curve would initially be very shallow. If very few of the thalli were broken until large forces were applied then the curve would be moved to the right.

When only the larger blades were used in the phase comparisons the graphs (Fig. 9,12) showed that at the same water velocities the gametophytes were more likely to be removed, at least in the exposed site for the June sampling time (Fig. 12). At a water velocity of 7 m/s, for example, between .1 and .2 of the tetrasporophyte population would be removed, whereas, between .2 and .7 of the gametophytes would be removed. A consistent difference of several percent between the phases could account for the difference in phase proportions observed. A larger sample size for gametophytes would probably separate the curves further and give a more precise estimate of the susceptibility of the phases to removal, but this graph at least demonstrates that a significant difference did exist. Of the population of Mazzaella lilacina in the exposed site in June 53% was over 15 cm long. Since the difference among the phases in susceptibility to removal only applied to the larger thalli the effect at the whole population level of having more gametophytes removed would likely be less than that suggested by Figure 4.12, however it still may be important.

The thalli most likely to be reproductive are the large ones. If more reproductive gametophytes are lost than reproductive tetrasporophytes the ability of the population to produce tetrasporophytes from spores would be decreased relative to its ability to produce gametophytes. In spite of this, exposed populations maintain the predominance of tetrasporophytes, suggesting that factors other than spore availability are responsible for the observed phase proportions.

The curves generated to test hypothesis 1 used a number of measurements from the population. Hypotheses 2-3 were tested to give insight into the curves by examining the phases for differences in stress to break, force to break, stipe-holdfast junction area, and blade area.

One might expect based on the different carrageenans present in gametophytes and tetrasporophytes that the two phases would have different material strengths. However, the second hypothesis, that the stress to break ($S_b$) for tetrasporophytes was different than that for gametophytes, was not accepted for the sheltered site or for the exposed site, even when only larger blades were used. Evidently, the free-living phases of Mazzaella lilacina have similar material strengths.

Stress to break can be further broken down into the force required to break the thalli and the cross-sectional area of the point of breakage, i.e. the stipe-holdfast junction. No differences were found between the phases in either of the above two measurements, although the differences in force to break were approaching significance in favour of the gametophytes in the sheltered site, when only large blades were used. In the exposed site, the difference was also nearly significant with the tetrasporophytes requiring more force to be broken.

Having larger blades would subject one phase to greater drag forces. In the absence of a difference in stress to break, as observed, the larger phase would be expected to be more susceptible to removal by waves. Only in the sheltered site when only large blades were measured did the difference in blade area between the phases approach significance. However, the difference was not great enough to make the gametophytes significantly more susceptible to removal by waves (fig. 9).

In the exposed site, using the large blades only, there were no significant differences between the phases in any of the variables measured, however, a greater proportion of the gametophyte population than tetrasporophyte population was removed at low water velocities (Fig. 12). The greater resistance to removal found in the tetrasporophyte population was due to a nearly significant difference in the forces to break the two phases, as well as to a slight trend towards larger blades among the gametophytes.

Since the curves generated for the probability of removal were very steep any separation between the bands for the gametophytes and tetrasporophytes translates into a large difference in the proportions of each phase which would be removed with water of a given velocity. Increased sampling may be sufficient to separate the confidence bands in the first three probability of removal vs. water velocity graphs.

A possibility which was not addressed in this study is that susceptibility to removal may vary over the course of an annual growth cycle with one or the other phase being more easily removed at different times. As well, no comparison was made separating vegetative and reproductive thalli. Further information about the timing, during the year and life cycle, of susceptibility to removal by waves would also be helpful for models of population dynamics.

## Conclusions

Several new insights have been gained into the spatial distribution of the gametophytes and tetrasporophytes of Mazzaella lilacina in the study areas. In the exposed site several times more tetraspores than carpospores were found in the population at all times of year suggesting that a majority of new blades found in clearings would be gametophytes; as was found for most clearing times in both sites. However, the phase proportions among recruited blades were never the same or even close to those found in the uncleared population in the exposed site. There were many more gametophytes in the recruits than in the surrounding population. In the sheltered site again there were more gametophytes among the recruits than among the surrounding thalli but the difference between the two was not great.

Based on the comparisons of the proportions of gametophytes among the recruits and the proportions of reproductive tetrasporophytes (parents) at the time clearings were made it appeared that gametophytes were better suited than tetrasporophytes for settlement and/or early growth in the sheltered site, and that tetrasporophytes were better suited than gametophytes for settlement and/or early growth in the exposed site.

In order for the population in the exposed site clearings to eventually reflect the surrounding population the mortality of gametophyte blades must be higher than the mortality of tetrasporophyte blades. Such a trend was observed, there was a slight increase in the proportion of tetrasporophytes in successive samplings of the exposed site clearings, suggesting that some post recruitment factors must be causing phase discriminate mortality. In the sheltered site the trend was towards an increased proportion of gametophytes.

One source of phase discriminate mortality was determined. It was shown that large gametophyte blades were more susceptible to wave induced mortality than large tetrasporophyte blades, in the exposed site. However, due to the small size of the new recruits it seems highly unlikely that the drag forces associated with moving water were responsible for the apparent mortality differences between the phases among the clearing populations.

Clearly, all of the major factors controlling the distributions of the gametophytes and tetrasporophytes of Mazzaella lilacina have not been determined.

Herbivory by micro- and macro- herbivores could be examined by seeding plates with a known density of spores of each phase, then culturing the plates in the presence and absence of various herbivores. As well, seeded plates could be grown in the field under various conditions in order to determine which post-recruitment stages are subject to the greatest mortality, and under what conditions.

References

Ang, P. Jr., R.E. De Wreede, F. Shaughnessy & L. Dyck (1990) A simulation model for an Iridaea splendens (Gigartinales, Rhodophyta) population in Vancouver, Canada. Hydrobiologia 204/205: 191-196

Bhattacharya, D. (1985) The demography of fronds of Chondrus crispus Stackhouse. J. Exp. Mar. Biol. Ecol. 91, 217-231

Buschmann, A.H. (1991) Amphipod food preference and Iridaea spp. (Rhodophyta) spore release and dispersal. J. Mar. Biol Ass. U.K. 71, 891-897

Buschmann, A.H. & P.A. Vergara (1993) Effect of rocky intertidal amphipods on algal recruitment: a field study. J. Phycol. 29, 154-159

Carrington, E. (1990) Drag and dislodgement of an intertidal macroalga: consequences of morphological variation in Mastocarpus papillatus Kutzing. J. Exp. Mar. Biol. Ecol. 139, 185-200

Chapman, A.R.O. (1986) Population and community ecology of seaweeds. Advances in Marine Biology 23,1-161

Craigie, J.S. & J.D. Pringle (1978) Spatial distribution of tetrasporophytes and gametophytes in four maritime populations of Chondrus crispus. Can. J. Bot. 56, 2910-2914

D'Antonio, C. (1986) Growth and reproduction of the red alga Rhodomela larix. Can. J. Bot. 64, 1499-1506

Denny, M.W. (1988) Biology and the Mechanics of the Wave-Swept environment. Princeton Univ. Press, Princeton, New Jersey, 320pp.

Destombe, C., J. Godin, Cl. Lefebvre, O. Dehorter, & Ph. Vernet (1992) Differences in dispersal abilities of haploid and diploid spores of Gracilaria verrucosa (Gracilariales, Rhodophyta). Botanica Marina 35, 93-98.

Destombe, C., J. Godin, M. Nocher, S. Richerd, & M. Valero (1993) Differences in response between haploid and diploid isomorphic phases of Gracilaria verrucosa (Rhodophyta: Gigartinales) exposed to artificial environmental conditions. Hydrobiologia 260/261, 131-137

De Wreede, R.E. & L.G. Green (1990) Patterns of gametophyte dominance of Iridaea splendens (Rhodophyta) in Vancouver Harbour, Vancouver, British Columbia, Canada. J. Applied Phycology 2: 27-34

De Wreede R.E. & T. Klinger (1988) Reproductive strategies in algae. In Plant Reproductive Ecology: Patterns and Strategies. Lovett-Doust, J. and Lovett-Doust, L. (Eds). Oxford University Press

Dudgeon, S.R. & A.S. Johnson (1992) Thick vs. thin: thallus morphology and tissue mechanics influence differential drag and dislodgement of two co-dominant seaweeds. J. Exp. Mar. Biol. Ecol. 165, 23-43

Dyck, L., R.E. De Wreede, D. Garbary (1985) Life history phases in Iridaea Cordata (Gigartinaceae): relative abundance and distribution from British Columbia to California. Jap. J. Phyc. 33: 225-232

Dyck, L. (1991) Some aspects of the demography of Iridaea splendens. M.Sc. Thesis, University of British Columbia

Garbary, D.J. and R.E. De Wreede (1988) Life-history phases in natural populations of Gagartinaceae (Rhodophyta): quantification using resorcinol. In Lobban C.S., Chapman, D.J., and Kremer, B.P. (Eds), Experimental Phycology: aA Laboratory Manual. Cambridge U.P.. Cambridge, 174-178

Green, L. (1989) MSc Thesis, University of British Columbia

Hannach, G. & B. Santelices (1985) Ecological differences between the isomorphic reproductive phases of two species of Iridaea (Rhodophyta: Gigartinales). Mar. Ecol. Prog. Ser. 22, 291-303

Hansen, J.E. (1977) Ecology and natural history of Iridaea cordata (Gigartinales, Rhodophyta) growth. J. Phycol. 13, 395-402

Hansen, J.E. & W.T. Doyle (1976) Ecology and natural history of Iridaea cordata (Rhodophyta: Gigartinales): population structure. J. Phycol. 12, 273-278

Hicks, C.R. (1982) Fundamental Concepts in the Design of Experiments, Third Edition. Holt, Rinehart, and Winston Inc.

Hurlburt, S.H. (1984) Pseusoreplication and the design of ecological field experiments. Ecol. Monogr. 52(2), 187-211

Koehl, M.A.R. (1986) Seaweeds in moving water: form and mechanical function. pp 603-634 In Givnish, T.J. (Ed). On The Economy Of Plant Form And Function. Cambridge Univ. Press., Cambridge. 717p.

Lazo, M.L., M. Greenwell & J. McLachlan (1989) Population structure of Chondrus crispus Stackhouse (Gigartinaceae, Rhodophyta) along the coast of Prince Edward Island, Canada: distribution of gametophytic and sporophytic fronds. J. Exp. Mar. Biol. Ecol. 126: 45-58

Littler, M.M. & D.S. Littler (1980) The evolution of thallus form and survival strategies in benthic marine macroalgae: field and laboratory tests of a functional form model. Am Nat. 116(1): 25-44

Luxoro, C. & Santelices, B. (1989) Additional evidence for ecological differences among isomorphic reproductive phases of Iridaea laminarioides (Rhodophyta: Gigartinales). J. Phyc. 25: 206-212

Mathieson, A.C. & R.L. Burns (1975) Ecological studies of economic red algae. V. Growth and reproduction of natural and harvested populations of Chondrus crispus Stackhouse in New Hampshire. J. Exp. Mar. Biol. Ecol. 17, 137-156

May, G. (1986) Life history variations in a predominantly gametophytic population of Iridaea cordata (Gigartinaceae, Rhodophyta). J. Phycol. 22: 448-455

McCandless, E.L., J.S. Craigie, & J.E. Hansen (1975) Carrageenans of gametangial and tetrasporangial stages of Iridaea cordata (Gigartinaceae). Can. J. Bot. 53, 2315-2318.

McCandless, E.L., J.A. West & M.D. Guiry (1983) Carrageenan patterns in the Gigartinaceae. Biochemical Systamatics and Ecology 11(3): 175-182

Melo, R.A. & M. Neushul (1993) Life history and reproductive potential of the agarophyte Gelidium robustum in California. Hydrobiologia 260/261, 223-229

Shaughnessy, F.J. & R.E. De Wreede (1991) Reliability of the resorcinol method for identifying isomorphic phases on the Gigartinaceae (Rhodophyta). J. Applied Phyc. 3: 121-127

Underwood, A.J. (1981) Structure of a rocky intertidal community in New South Wales: patterns of vetical distribution and seasonal change. J. Exp. Mar. Biol. Ecol. 51, 57-85

Underwood, A.J. & E.J. Denley (1984) Paradigms, explanations, and generalizations in models of the structure of intertidal communities on recky shores. In Ecological Communities: Conceptual Issues and the Evidence. Strong, D.R.Jr., D. Simberloff, L.G. Abele, & A.B. Thistle (Eds). Princeton University Press, Princeton.

Vadas, R.L.Sr., S. Johnson, and T.A. Norton (1992) Recruitment and mortality of early post-settlement stages of benthic algae. Br. Phyc. J. 27, 331-335

Vogel S. (1989) Drag and reconfiguration of broad leaves in high winds. J. Exp. Biol. 40(217), 941-948

Waaland, J.R. (1973) Experimental studies on the marine algae Iridaea and Gigartina. J. Exp. Mar. Biol. Ecol. 11, 71-80

Wilkinson, L. (1990) Systat: The System for Statistics. Evanston, IL: Systat, Inc.