EFFECTS OF SHORT-TERM LIGHT REDUCTION DURING DIFFERENT SEASONS ON SURVIVAL OF INTERTIDAL AND SUBTIDAL EELGRASS, ZOSTERA MARINA, IN PADILLA BAY, WASHINGTON

Douglas A. Bulthuis

March 1996

Final Report for
NOAA/SRD Award # NA27OR0216

Padilla Bay National Estuarine Research Reserve
Shorelands and Water Resources Program
Washington State Department of Ecology

1043 Bayview-Edison Road
Mt. Vernon, WA 98273

Table of Contents

List of Tables

List of Figures

ABSTRACT

The purpose of this study was to investigate the seasonal effects of short-term light reduction on survival of the eelgrass, Zostera marina. Light was reduced to 10% of ambient photosynthetically active radiation for periods of 8-10 weeks in 1.25 m by 1.25 m plots in field experiments conducted on intertidal and subtidal populations of Z. marina. Experiments were conducted in summer, autumn, winter and spring. Delayed effects and recovery were monitored for 6-18 months after treatment. Two months of light reduction resulted in reduced density of Z. marina during spring and summer at both the intertidal and subtidal sites. No significant decreases compared to controls were observed in autumn and winter during the two months of light reduction, but delayed effects on density were observed in some treatments during subsequent seasons of growth. Very little recovery was observed in any of the plots during the 6-18 months following treatment. This study indicates that light reduction may have a greater effect on survival of Zostera marina during spring and summer than it does in autumn or winter; that light reduction during autumn and winter may have delayed effects on density; that intertidal Z. marina may be more sensitive to light reduction than subtidal Z. marina; and that recovery from even short-term light reduction may take more than one year.

INTRODUCTION

Seagrasses are the basis of important estuarine and shallow coastal communities because of their high productivity (Zieman & Wetzel 1980), their role in structuring the environment (Stoner & Lewis 1985), in providing habitat for fish (Pollard 1984), and other marine animals (Bell *et al.* 1984), in nutrient cycling (Short 1983), and stabilization of the sediment (Harlin *et al.* 1982, Bulthuis *et al.* 1984). The importance of seagrasses in the Padilla Bay estuary has been demonstrated by studies of the area covered by seagrasses (Webber *et al.* 1987, Bulthuis 1991, 1995), their productivity (Thom 1990), their role in nutrient cycling (Williams & Ruckleshaus 1993), and in supporting crabs (Dinnel *et al.* 1993, McMillan *et al.* 1995) and food organisms of juvenile fish (Simenstad *et al.* 1988, 1995).

However, seagrass communities are not stable (Larkum & West 1983), and human induced changes to the marine and estuarine environment have caused major losses of seagrasses (Orth & Moore 1983, Cambridge & McComb 1984, Shepherd *et al.* 1989, Giesen *et al.* 1990). Two of the major factors implicated in loss of seagrasses in the above studies are increased nutrient inputs to marine coastal areas and increased turbidity of the water (Kemp *et al.* 1983, Cambridge *et al.* 1986, Shepherd *et al.* 1989, Giesen *et al.* 1990). Nutrient enrichment of coastal marine and estuarine waters results in increased phytoplankton, epiphytes on leaves of seagrasses (Silberstein *et al.* 1986, Williams & Ruckelshaus 1993) and development of macroalgae (Harlin & Thorne-Miller 1981, Perkins & Abbott 1972, Bach & Josselyns 1978, Thom & Albright 1990). Increased macroalgal biomass, particularly of species in the genera Ulva and Enteromorpha, is one of the most frequently reported results of eutrophication (Bulthuis & Cowdell 1982). This biomass can accumulate in large mats which may wash onto beaches or settle in intertidal areas. When such mats of macroalgae settle onto intertidal areas inhabitated by seagrass, they reduce light to the seagrasses and can cause anaerobic conditions underneath the mat (Perkins & Abbott 1972) and have been reported as the cause of seagrass dieback (Den Hartog 1994).

Macroalgal biomass fluctuates seasonally, particularly in response to increased nutrient inputs (Thom 1980, Thorne-Miller *et al.* 1983, Thom & Albright 1990). Macroalgal mats may be shifted from place to place in an estuary by waves and currents, first covering an eelgrass bed and then leaving it uncovered. Similarly, turbidity fluctuates widely in estuaries caused in part by changes in river flow or human activities in the watershed. Thus, macroalgal mats and turbidity peaks may affect seagrasses for only short periods of time during the year rather than continuously. One of the objectives of this study is to investigate the effects of short-term light reduction on survival of the eelgrass, Zostera marina.

Padilla Bay contains one of the most extensive stands of seagrasses in the Pacific Northwest (Phillips 1984, Bulthuis 1995). Most of the lower intertidal and upper subtidal in Padilla Bay is covered by the seagrass Zostera marina where growth and morphology indicate healthy stands (Bulthuis 1995). However, the lower limit of distribution of Zostera marina in Padilla Bay is only about -3.0 m (Bulthuis, unpublished observations) which is not nearly as deep as the -6.6 m that Phillips' work indicated was the light compensation depth for eelgrass survival in other parts of Puget Sound (Phillips 1972, 1974). The more restricted depth distribution in Padilla Bay than that which Phillips found for Puget Sound as a whole is probably caused by decreased water clarity in Padilla Bay. The seagrasses in Padilla Bay cover a large area that has a very gradual slope from the upper intertidal to the subtidal (Bulthuis 1991). Because of the very gradual slope an increase in turbidity and a decrease in the light compensation depth of one or two meters could result in the loss of an extensive area of seagrass habitat.

Studies on the importance of light to seagrasses have usually emphasized the lower limit of distribution which is often controlled by light (Duarte 1991). Light can also be an important factor in the intertidal because higher temperatures result in an increased minimum light requirement (Bulthuis 1983b, 1987). In Western Port, Australia, increased suspended sediment in the water was accompanied by death and loss of intertidal seagrasses at the same time as subtidal seagrasses survived (Shepherd *et al.* 1989). Therefore, a decrease in light may affect intertidal seagrasses without necessarily increasing the light compensation depth in a progressive and linear fashion. Intertidal seagrasses also are located closer to the source of suspended sediments to Padilla Bay and subjected to floating mats of green algae. Therefore, in this study, light reduction experiments were conducted on both a subtidal and intertidal population of Zostera marina in Padilla Bay.

Seagrasses and light reduction

Seagrasses have died back from large embayments and estuaries throughout the world and often a major cause for the loss was decreased light from either increased nutrients or increased suspended solids. In Cockburn Sound, Western Australia, the die back of seagrasses was attributed to increased nutrients. These nutrients caused an increase in phytoplankton and epiphytes which in turn caused a decrease in light reaching seagrass leaves. This resulted in death of the seagrasses and loss of about 3,300 hectares of seagrass beds (Cambridge & McComb 1984, Cambridge *et al.* 1986, Silberstein *et al.* 1986, Shepherd *et al.* 1989). In Chesapeake Bay, Orth and Moore (1983) documented a huge loss of seagrasses and suggested that "factors affecting the quantity and quality of light reaching the plant surface" may be related to the decline of seagrasses. Similarly, Kemp *et al.* (1983) concluded that the relative contribution of nutrients>sediments>herbicides as factors causing the decline of submerged aquatic vascular plants in Chesapeake Bay. Nutrients and sediments affected plants through a reduction in light. In Gulf St. Vincent, South Australia, seagrass declines were reported near sewage outfalls where nutrient-induced epiphyte loading reduced the light reaching seagrass leaves (Shepherd *et al.* 1989). In Western Port, Victoria, Australia the loss of 17,800 hectares was attributed to increased suspended sediments settling on seagrass leaves and reducing light (Shepherd *et al.* 1989). In these and other studies of seagrass losses, a reduction in light to seagrasses (often related to increased suspended sediments or increased nutrient inputs) has been suggested as one of the major causes (Phillips 1980, Livingston 1987, Shepherd *et al.* 1989).

The importance of light to growth and survival of seagrasses has been demonstrated in numerous studies which indicate that the lower limit of distribution of seagrasses is often controlled by light (Backman & Barilotti 1976, Bulthuis 1983a, Dennison 1987, Dawes & Tomasko 1988, Orth & Moore 1988, Duarte 1991). Several studies have examined the effects of reduced light on Zostera marina. Dennison and Alberte (1985) recorded decreased biomass and leaf growth rates when Zostera marina was shaded for one month and cellular photosynthetic changes during one of their two experimental periods. Their work demonstrated the importance of the hours per day that light intensities saturated Zostera marina photosynthesis rather than average light intensity alone (Dennison & Alberte 1982, 1985, Dennison 1987). Backman & Barilotti (1976) decreased light over Zostera marina and demonstrated a reduction in standing crop in a seagrass bed near the lower limit of distribution. In other seagrasses, Neverauskas (1988) decreased light by 50% over Posidonia sinuosa and Posidonia angustifolia. There was no change for six months, but a rapid decline over the following six months. Congdon and McComb (1979) reduced light over Ruppia maritima and recorded reduced densities and standing crop over the course of a year. When irradiance was reduced to Heterozostera tasmanica, density decreased, leaf length increased and leaf width and leaf growth rate remained the same (Bulthuis 1983a). Two of the questions that the above studies have not addressed are: "Are there seasonal differences in the effects of light reduction on Zostera marina?" and "Are there longer term (one year) delayed effects of short term (one to two months) reductions in the light to Zostera marina?".

Seasonal differences in effects of light reduction

The data on seasonal effects of light reduction on Zostera marina is sparse and conflicting. Some seasonal differences in the effects of light reduction may be expected. When light was reduced to the related seagrass, Heterozostera tasmanica, density decreased more rapidly during summer than during winter (Bulthuis 1983a). These results with Heterozostera tasmanica in Western Port, Australia, indicate that Zostera marina in Padilla Bay would be more susceptible to reductions in light in summer than in winter. However, the seasonal pattern of growth of Heterozostera tasmanica and Zostera marina differ somewhat (cf Bulthuis & Woelkerling 1983 with Phillips 1972, Sand Jensen 1975, Nienhuis & deBres 1980, and Thom 1990) and the effects of light reduction differed somewhat particularly with regard to leaf growth rate (cf Bulthuis 1983a with Dennison & Alberte 1985). Dennison (1987) indicated that individual seedlings of Zostera marina in Great Harbor, Massachusetts may penetrate deeper than the perennial seagrass meadow during summer, but die back during subsequent months because of low light levels. Dennison's study indicates that Zostera marina in Padilla Bay would be more susceptible to reductions in light in winter than in summer. Thus, the available data regarding seasonal differences in the effects of light reduction on Zostera marina is sparse and conflicting as to whether eelgrass would be more susceptible to reduction in light in winter or in summer.

Delayed effects of light reduction

Long term or delayed effects may be expected from short term light reduction. A few observations on long term effects have been made in studies whose focus had been the immediate effects of light reduction. Backman & Barilotti (1976) removed light reduction screens from three plots after two months and found that recovery took one to six months. Dennison & Alberte (1985) reported that all seagrasses (Z. marina) died several weeks after removing the 30 day light reduction screen from one station (deep), but that full recovery occurred within 14 days at a second station (shallow). Seasonal differences may be expected if productivity in one season (e.g. spring and summer) is used to support growth during a less favorable season (e.g. winter). Delayed effects may be expected if short-term reductions in light intensity reduce the photosynthate exported and stored in the rhizomes. With less energy reserves in the rhizome, seagrasses may be unable to survive the following winter and spring. Thus, seagrasses may continue to survive through the late summer and autumn, even if light had been reduced in early summer, but the seagrass may fail to produce new leaves the following year because of reduced overwintering rhizome biomass.. Thus, another objective of the present study was to determine whether there were any delayed effects to short-term (two months) light reduction.

The objectives of the present study have been to investigate the effects of short-term (two months) light reduction on survival of eelgrass, Zostera marina, in Padilla Bay, Washington. The specific factors that have been investigated are seasonal differences in the effects of short-term light reduction, differences between an intertidal and a subtidal populations of Z. marina, delayed effects of short-term light reduction, and recovery during the first year following short-term light reduction.

METHODS

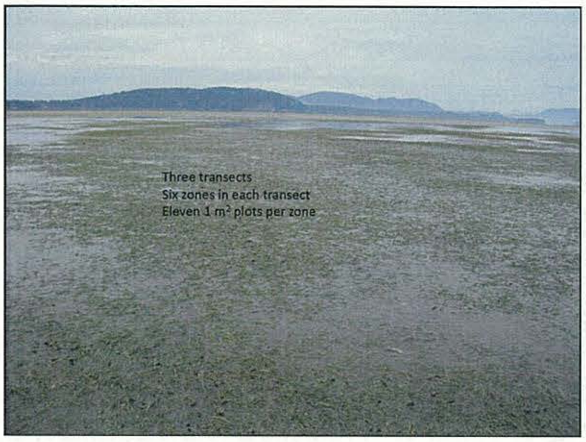
Light available to Zostera marina was reduced experimentally to 10% of control irradiance using neutral density screens at two sites: an intertidal site and a subtidal site (Fig. 1). The intertidal site, approximately 25 m by 25 m, was located in an apparently uniform bed of Z. marina that extended more than 100 m in all directions. Within the uniform bed, but outside the experimental site were bare patches varying in size from about 2 m diameter to 10 m diameter. Sediment level was about 0 to +0.25 m depth where 0 is chart datum, MLLW. Spring tidal amplitude is about 2.5 m. The subtidal site was located at a depth of about -2.5 m, about 150 m above the lower limit of distribution on a gently sloping bottom. Zostera marina extended more than 100 m in all directions with some evidence of thinning Z. marina close to the lower limit. The subtidal site was a rectangle about 12 m wide and 100 m long oriented parallel to the lower limit of distribution so that the whole experimental area was at a similar depth and similar distance from the lower limit of distribution.

Experiments were initiated in March, June, September, and December 1993. An additional experiment was initiated in March 1994, in which only density of Z. marina was measured. At each site four replicates of three "treatments" were allocated (in a random block design) to plots in three rows and four columns for each experiment. Subsequent experiments were allocated in an adjacent block of rows and columns. Plots were 1 m by 1 m within a 1.25 m by 1.25 m screen (light reduction plots) or a 1.25 m by 1.25 m frame without screen (control plots). The space between plots and between rows was at least 2 m. The three treatments were control, light reduction, and pretreatment. Biomass samples were taken from the pretreatment plots at the time each experiment was started, with no further samples or measurements taken from pretreatment plots. At the end of the two months of light reduction, biomass samples were taken from one control and one light reduction plot. Chlorophyll concentration in the leaves of Z. marina was determined in four control and four light reduction plots at the beginning of each experiment, and in three control and three light reduction plots after one month and after two months of light reduction. Density was determined in each plot at the beginning of the experiment and monthly in each control and light reduction plot (except for the ones from which biomass samples were taken) thereafter through December 1994, 21 months after initiation of the first experiment.

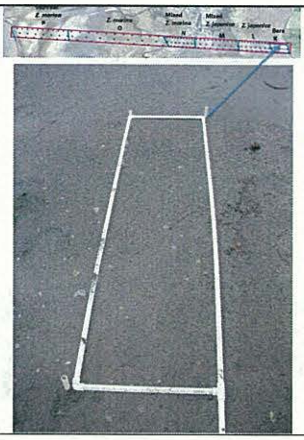
Screens were set up on PVC frames 0.55 m above the sediment at the intertidal site and 0.85 m above the sediment at the subtidal site so as not to restrict water flow. Screens were made of three layers of neutral density greenhouse shade cloth. Screens were cleaned each month. Light measurements with a PAR (photosynthetically active radiation, 400 - 700 nm) light sensor made under the screens and outside of the screens both during submergence and emergence indicated an average light reduction of about ten percent with a range of 4 - 23%.

All measurements and sample collection at the subtidal site were conducted with SCUBA. At the intertidal site, SCUBA was used during autumn and winter, but during late spring and summer measurements at the intertidal site were made by walking to the site during suitable daytime low tides.

Biomass was measured in triplicate quadrats (0.25 m by 0.25 m) in each plot. Above ground leaf material was clipped at the sediment surface and placed in marked bags, kept on ice for transport to the laboratory, stored at 5 °C for up to seven days, washed in freshwater, and cleaned of debris and large epiphytes. Sediments to a depth of 0.15 m were dug from each biomass quadrat, rinsed of sediment in the field, transported and stored as for leaf material, and cleaned of shells and animals in freshwater. Dry weight



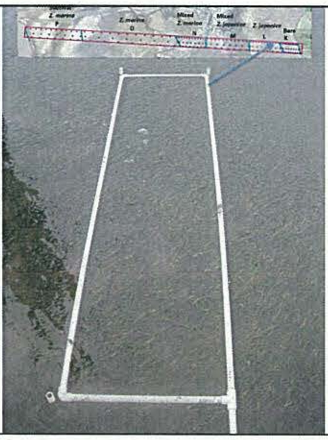
Three transects
Six zones in each transect
Eleven 1 m^2 plots per zone



Plot K4

(Bare)

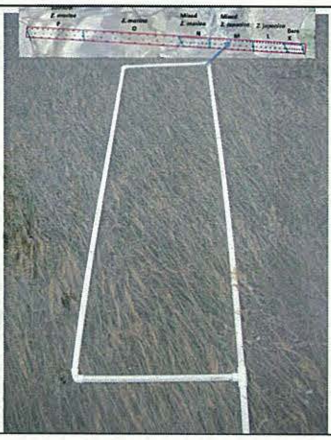
July 2011



Plot L2

Z. japonica

July 2011



Plot M6

Mixed
Z. japonica

July 2011

Plot N5

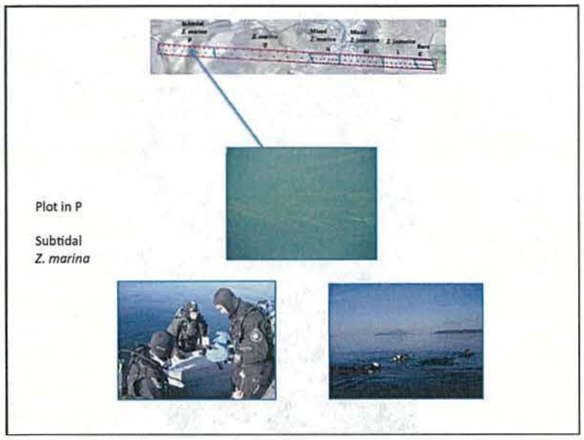
Mixed
Z. marina

July 2011

Plot O6

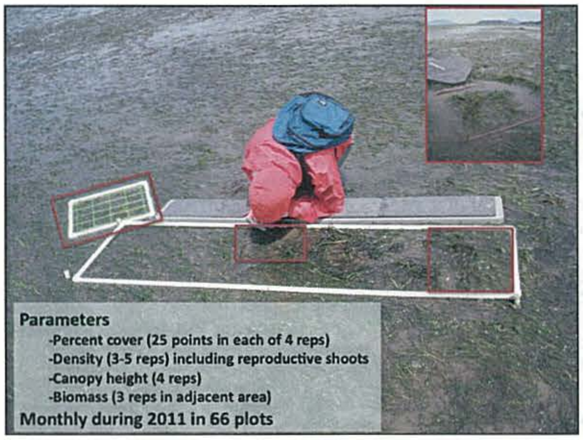
Intertidal
Z. marina

July 2011



Plot in P

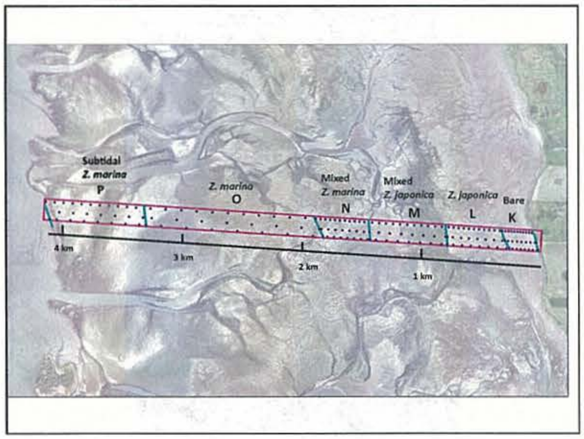
Subtidal
Z. marina

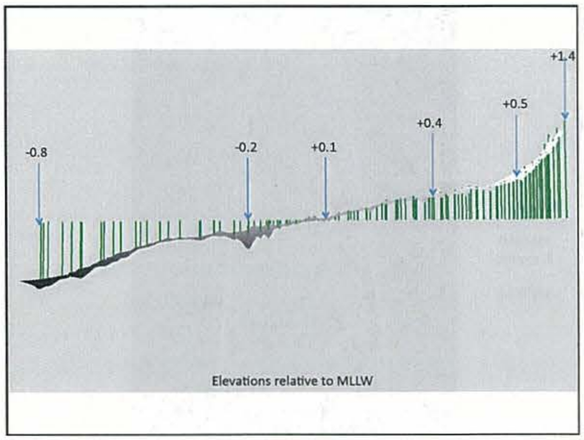


Parameters

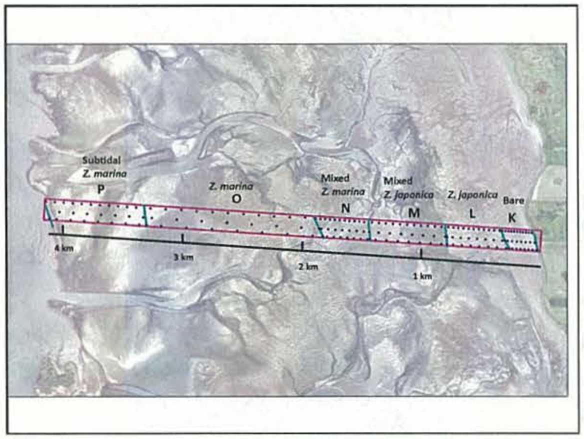
- -Percent cover (25 points in each of 4 reps)
- -Density (3-5 reps) including reproductive shoots
- -Canopy height (4 reps)
- -Biomass (3 reps in adjacent area)

Monthly during 2011 in 66 plots





Elevations relative to MLLW



of leaf and root/rhizomes was determined by drying to a constant weight at 105 °C; ash-free dry weight by heating to 550 °C for two hours and reweighing.

Chlorophyll was measured in 1 cm^2 sections from the center of 2-4 whole leaves haphazardly selected in randomly allocated quadrats in each plot. Leaf samples were macerated, extracted in 90% acetone overnight (Dennison 1990) and absorption measured in a narrow bandwidth spectrophotometer and chlorophyll a and b calculated with the equations of Jeffrey and Humphrey (1975).

Density was measured non destructively by counting all shoots above the sediment surface in 0.25 m by 0.25 m quadrats, three randomly allocated quadrats per plot at the intertidal site and four per plot at the subtidal site.

RESULTS

The experimental frames were designed to reduce light to the eelgrass and to have little or no effect on other factors affecting growth and survival of Z. marina. However, drifting algae tended to get caught on the frames at the intertidal site in both control plots and light reduction plots and lay on the eelgrass when the tide flats were exposed. Each month these drift algae were removed from the frames. The affected area was usually within 0.1 m of the corner posts and thus not in the measured plot area (central 1 m^2). But in May 1993, accumulations of macroalgae (mainly of the genera Laminaria, Ulva, and Enteromorpha) were much larger than other months and covered parts of the plot area in both control plots and light reduction plots. At the time, the spring (March 1993) experiment was the only experiment set up. Similar accumulations of macroalgae were not seen during the remainder of the study. Thus, eelgrasses in the spring (March 1993) intertidal experiment experienced both reduced light with screens in the treatment plots and reduced light (and other effects of macroalgal mats) in both the treatment and control plots.

During the 21 months that eelgrass density was measured at the experimental sites, changes were noted in the patchiness of the intertidal site. One of the "sparse-bare" patches in the surrounding meadow increased in size during the summer of 1993 and extended into a few of the plots (both control and light reduction plots) in the spring 1993 experimental area. Thus, results measuring 'delayed effects' and 'recovery' of the spring 1993 experiment are interpreted with caution.

Seasonal fluctuations

Density, leaf dry weight and chlorophyll content of the leaves of Zostera marina were measured in control plots on numerous occasions throughout the year as each experiment was set up and concluded. Combining these data from the control plots provides an indication of the natural seasonal changes in these parameters. Density was measured in control plots from the different seasonal experiments each month. The density was similar in control plots from all experiments except for the intertidal spring 1993 experiment. Control plots from spring 1993 had lower density in June and had less than control plots from other seasons almost every month for the following 18 months. Therefore, the spring 1993 controls were not included after June 1993 when computing the mean densities for all controls at the intertidal site.

Density of Z. marina at the subtidal site was highest in summer and autumn with 100-140 shoots per m^{-2} and lower in winter and spring with 50-80 shoots per m^{-2} (Fig. 2). Peak density during 1994 was about 70% as high as peak density during 1993. Density at the intertidal site was always higher than density at the subtidal site (Fig. 3), but the seasonal pattern at the intertidal site was almost the reverse of the subtidal site:

density increased during winter to a maximum during spring and then decreased during late autumn (1993) or summer and autumn (1994). Peak densities during spring at the intertidal site were about two times the densities during autumn (Fig. 3).

The dry weight of leaves per unit area of Z. marina at the intertidal site was at a maximum in summer, after the spring maximum in density, and at a minimum in winter of about one-fourth to one-third of the summer maximum (Fig. 4). At the subtidal site, the seasonal pattern of dry weight of leaves was similar to the intertidal site, but summer maxima were about twice as high (Fig. 5). Ash-free dry weight averaged 55% of leaf dry weight at the intertidal and subtidal sites. Percent ash-free dry weight was only four to five percent below the mean during the summer months, so the seasonal pattern of ash-free leaf dry weight was similar to the pattern for leaf dry weight.

Leaf dry weight per individual shoot at the intertidal site indicated two sizes of shoots: one during May to September with a weight of 0.4 to 0.5 g per shoot, and a second during December to March with a weight of 0.1 to 0.2 g per shoot (Fig. 6). At the subtidal site, leaf dry weight per shoot was always greater than at the intertidal site (from 0.4 to 2.2 g per shoot) and there was some indication of a seasonal trend with a maximum in summer and a minimum in winter/early spring (Fig. 7).

The concentration of chlorophyll a and b in the leaves of Zostera marina was similar throughout the year at both the subtidal and intertidal sites with some indication of an increase in chlorophyll in winter (December to March) at the intertidal site. At the subtidal site, monthly mean concentrations were usually 25-32 µg per cm^2 and at the intertidal site 19-23 µg per cm^2 from March through October and 26-33 µg per cm^2 from

November to March. Mean concentrations at the subtidal site were about 5 µg per cm^2 higher than at the intertidal site.

The photon flux density at the water surface in Padilla Bay fluctuated throughout the year as expected for this latitude (48°N) with a maxima in June and July and a minima in December and January (Fig. 8). Water temperature in the channels in Padilla Bay fluctuated between a high of 15 °C in July and August to a low of 5 °C in during December to February (Fig. 8). Temperature on the intertidal flats during low tide can be several degrees higher or lower depending on air temperature, wind, and solar radiation.

Immediate effects of light reduction

At the subtidal site, light reduction during spring 1993 caused a reduction in density of about 25%, but this occurred at a time when control plot densities nearly doubled (Fig. 9). Leaf dry weight in control plots quadrupled from March 25 to June 2, 1993 during which time leaf dry weight in light reduction plots decreased to less than one-fourth of the initial weight (Table 1). Similarly, ash-free dry weight of leaves, dry weight and ash-free dry weight of rhizomes, and leaf dry weight per shoot all increased in control plots and either decreased or remained the same (leaf dry weight per shoot) in the light reduction plots (Table 1). On the other hand, the concentration and ratio of chlorophyll in the leaves of Z. marina did not change with time or with reduced light (Table 2). During spring 1994 density in control plots appeared to increase slightly, whereas density of Z. marina in light reduction plots decreased to less than half in two months (Fig. 9).

At the intertidal sites, light reduction for two months during spring caused an immediate and drastic reduction in density of Z. marina both in 1993 and 1994 (Fig. 10) at a time when density in control plots were changing very little if at all. Similarly, the dry weight and ash-free dry weight of leaves and rhizomes were much lower after two months of light reduction compared to controls (Table 3). Even the few surviving shoots of Z. marina in the light reduction plots had about one half the leaf dry weight per shoot as did shoots in control plots (Table 3). Light reduction had no measurable effect on chlorophyll a, chlorophyll b, or their ratio in leaves of Z. marina during the spring experiment at the intertidal site (Table 4).

During summer, at the subtidal site, density of Z. marina decreased during two months of light reduction to about one half of density in control plots (Fig. 9). Similarly, dry weight of leaves and leaf dry weight per shoot deceased in light reduction plots (Table 1). Nine weeks after treatment the concentration of chlorophyll in leaves of Z. marina under light reduction screens was lower than in control plots (Table 2).

At the intertidal site, during summer, the response of Z. marina to light reduction was similar to the response at the subtidal site (Fig. 10, Table 3) except that there was no significant decrease in chlorophyll per unit leaf area, with even some indication of an increase in chlorophyll after four weeks (Table 4).

During autumn at the subtidal site, density decreased similarly in both control plots and light reduction plots so that there was no measurable effect of light reduction on density of Z. marina (Fig. 9). In the two plots in which leaf dry weights were measured, there was less in the light reduction plot than in the control plot (Table 1). Chlorophyll content of leaves of Z. marina was lower in light reduction plots than in control plots after four weeks, but similar in concentration after twelve weeks (Table 2). There was no change in chlorophyll a/b.

During autumn at the intertidal site, light reduction had no measurable effect on density with decreases measured in both control plots and light reduction plots (Fig. 10). Leaf dry weight and leaf dry weight per shoot, on the other hand, were lower in the single light reduction plot compared to the single control plot (Table 3). Chlorophyll a and b appeared to increase after four weeks and nine weeks of light reduction, but there was no effect of chlorophyll a/b (Table 4).

During winter at the subtidal site, light reduction had no measurable effect on density, leaf dry weight, leaf dry weight per shoot, or chlorophyll a/b of Z. marina (Fig. 9, Tables 1 and 2). After ten weeks of light reduction the chlorophyll content was lower in leaves from light reduction plots than in control plots (Table 2).

During winter at the intertidal site, light reduction for eleven weeks had no measurable effect on density, leaf dry weight, leaf dry weight per shoot, chlorophyll content of leaves or chlorophyll a/b of Z. marina (Fig. 10, Tables 3 and 4).

Delayed effects and recovery after light reduction

After removal of the screens that reduced light for ten weeks during spring 1993 at the subtidal site, density of Z. marina continued to decline through all seasons (Fig. 11). Sixteen months after removal of the screens, the lowest density in the treatment plots was recorded; and although plots were monitored for nineteen months after the ten week light reduction, the plots never recovered their previous density, nor the density of adjacent control plots (Fig. 11). After removal of the screens after eight weeks in spring 1994, density of Z. marina at the subtidal site increased parallel to the increases in the control plots. In the seven months following removal of the screens, Z. marina did not decline further (as measured by density of shoots), nor did it recover to densities recorded in the control plots (Fig. 12).

At the intertidal site, after removal of the screens, only a few scattered plants remained in the light reduction plots. These few plants remained during the following nineteen months with reported means fluctuating widely as randomly allocated quadrats included or excluded the few plants (Fig. 13). Z. marina never recovered in these plots, but control plots in this area also declined over nineteen months (Fig. 14). The area in which these plots were located appeared to be undergoing a change during this period of time (see second paragraph of Results section). In spring 1994, after removal of screens, density of Z. marina remained low and neither declined further nor recovered during the following seven months (Fig. 15).

Following light reduction during summer, Z. marina in subtidal plots never recovered during the following fifteen months (Fig. 16). Delayed effects were not obvious, but during the second growing season the difference between control and light reduction plots was greater than during the first season (Table 5, Fig. 16).

In intertidal plots, after light reduction during summer, no further decline was observed during the following sixteen months although density did not increase in light reduction plots the following spring when density in control plots tripled (Fig. 17). There was no clear sign of recovery during this period, although density in control plots declined during the winter of 1994 to values similar to the densities observed in the summer light reduction plots (Fig. 17).

Light reduction during autumn at the subtidal site did not cause a decrease in density different from the control plots (Fig. 9). During the following twelve months, densities in the light reduction plots fluctuated similar to the control plots, indicating no delayed effects (Fig. 18).

In contrast, at the intertidal site, where light reduction during autumn also had not caused an immediate decrease in density (Fig. 10), there was a clear delayed effect as density of Z. marina continued to decline in plots that had had light reduced (Fig. 19). At the same time density in control plots increased dramatically during winter and spring (Fig. 19). By the time of the second winter, thirteen months after the light reduction screens had been removed, density in the light reduction plots was similar to density in the control plots. Whether these plots fully recovered is not known, because of the termination of the experiment. Full recovery would be indicated by a large increase in density during the following winter and spring (1995) as occurred during 1993 and 1994 in the control plots (Fig. 19).

During winter at the subtidal site, light reduction had not caused a significant decrease in density (Fig. 9). During the next two and a half months density in light reduction plots was slightly lower than in control plots, but in May 1994 there was still not a clear difference between control and light reduction plots (Fig. 20). During the next three months of summer, density in the control plots almost doubled while density in light reduction plots decreased slightly (Fig. 20). Thus, a delayed effect from light reduction during winter was observed the following summer, four and more months after the light reduction screens had been removed.

At the intertidal site, also, light reduction during winter had not caused a significant decrease in density (Fig. 10). During the following nine months density in the light reduction plots was slightly lower than in the control plots, but the effect on density was equivocal and difficult to interpret with confidence (Fig. 21). By December 1994, the control plots appeared to be increasing while the treatment plots continued to decline (Fig. 21), possibly indicating a delayed effect.

DISCUSSION

Short-term (8 - 12 week) light reduction (10%) caused a decrease in density of Zostera marina at both a subtidal and intertidal site during spring and summer, but not during winter or autumn (Table 5). Zostera marina was more sensitive to light reduction in spring and summer than during winter. These results are counter intuitive because there is greater incident light during summer than winter (Fig. 8). Indeed, the seasonal changes in incident light have been invoked by many authors as causes for the seasonal changes in biomass and density of eelgrasses (Sand-Jensen 1975, Kentula and McIntire 1986, Dennison 1987, Thom and Albright 1990, Thom 1990).

If reduced light during winter is causing the seasonal decrease in biomass and density, a further reduction during winter, such as caused by the light reduction screens, would have been expected to cause a further decrease in density. The lack of any decrease in density under screens indicates that the seasonal decrease in density of Z. marina is not caused primarily by reduced solar photon flux density during winter. Light may be a secondary factor, interacting with other factors such as temperature and endogenous seasonal patterns, but these results indicate that light is not the major controlling factor causing reduced density of Z. marina during winter in Puget Sound.

Short-term (8 - 10 week) light reduction (10%) had a greater effect on Z. marina at the intertidal site than at the subtidal site: the light reduction screens caused a greater decrease in density in spring and summer at the intertidal site than at the subtidal site (Figs 9 and 10). Again, these results are counter intuitive because there is greater incident light at the intertidal site than at the subtidal site. The increased effect of light reduction at the intertidal site may have been due (among other things) to differences in temperature or differences in the suitability of the site for growth of Z. marina. Temperature at the two sites was not measured during this study, but would be expected to be similar. Tidal flow into Padilla Bay tends to bring water from the area of the subtidal site to the area of the intertidal site with each daily tide (Bulthuis and Conrad 1995) and therefore temperatures should be similar. Temperatures taken at stations near the experimental sites during a water quality study during 1985-1986 (Cassidy and McKeen 1986) indicated mean temperatures within 1 - 2 °C of each other all seasons of the year. These temperatures, however, were taken during high tide when the sampling vessel could access the intertidal site. During low tides the temperature of the ponded water at the intertidal site may increase up to 10 °C higher than during high tide (Bulthuis, unpublished data). Higher temperatures increase the respiration rate (Bulthuis 1983b, Marsh *et al.* 1986) of seagrasses and thereby increase the minimum light required for a positive carbon balance (Bulthuis 1987). Therefore, the greater effect of light reduction at the intertidal site could be a result of a greater light requirement because of higher temperatures. This may be a partial explanation of some of the differences between the subtidal and intertidal sites, but the length of time of temperature increase during low tide seems too short to explain the substantial differences in response to light reduction, particularly during spring and autumn.

A second "explanation" for the increased effect of light reduction at the intertidal site is the overall marginal habitat that such a site represents for Z. marina. Z. marina is essentially a submerged marine plant that is able to extend its range up into the intertidal when the periods of exposure are short, or the topography of the sand/mud flat forms a shallow (1 - 5 cm deep) bowl that retains water during low tide, or the morphology of the shoots and leaves retards water flow off of the flats (Powell & Shaffner 1991), or the leaves lie directly on the wet sediment surface. All of these situations keep the leaves wet in an environment where the leaves are in danger of drying out and dying. Other factors, such as freezing, ice scour, and grazing by herbivorous birds (Baldwin & Lovvorn 1994) also make the intertidal habitat marginal for growth of Z. marina. In such an environment, the additional stress of lowered light may have more effect than in a suitable subtidal habitat. That is, plants in the intertidal that may have multiple stressors may not have been able to build up the reserves (in rhizomes or shoots) to survive an 8 - 12 week reduction in photon flux density to 10% of control irradiance.

A third objective of this study was to investigate the delayed effects of short-term light reduction; that is, "Are there further changes in the Z. marina population within experimental plots during the following twelve months of full light that may be attributable to the short-term light reduction?" The only experiments that showed a clear delayed effect were the winter experiment at the subtidal site and the autumn experiment at the intertidal site. In addition, there was a slow continuous decline in density after removal of the screens in spring at the subtidal site. The delayed effects of the light reduction during winter at the subtidal site were especially striking because the treatment plots and control plots had about the same density for three months following removal of the screens and then during summer density in the control plots increased while density in treatment plots remained the same (Fig. 20). Thus, the effects were first observed about four months after the screens had been removed. At the end of the autumn intertidal experiment, densities in the control plots and the light reduction plots were similar. However, during the next three months (December to February) density in the control plots increased three to four times the November minimum while density in the light reduction plots remained similar or increased only slightly (Fig. 19). One explanation for the delayed effects of light reduction may be reduced carbohydrate reserves in the rhizomes. The reserves may have been used to sustain shoot growth during the time of light reduction so that the total density of shoots did not decrease. However, during the time of density increase (winter for intertidal, summer for subtidal) there were insufficient reserves in the light reduction plots to produce new shoot growth. As a result, density would remain constant in the light reduction plots while it increased in the control plots. Dennison & Alberte (1985) also reported delayed effects of light reduction. They reduced light for 30 days to Z. marina at a "deep" station and several weeks after they removed the screen, all Z. marina died. However, the present study is the first documentation of delayed effects first being observed some three months after the light reduction.

A fourth objective of this study was to investigate the rate of recovery during the year following short-term light reduction. Assessment of "recovery" will depend on the definition of what constitutes recovery. When "recovery" is defined as similar shoot density for two consecutive months in control and light reduction plots, then recovery was observed in the intertidal summer and autumn experiments and in the subtidal winter experiment (Figs. 17, 19, 20). However, in all three cases, "recovery" occurred because the density in control plots declined to the values observed in the light reduction plots, not because the density in the light reduction plots increased. If "recovery" is defined as an increase in shoot density at the time when density increased in control plots (summer for subtidal, winter for intertidal) then no "recovery" was observed in any of the experiments during the 7-19 months following removal of the screens (Table 5). Backman and Barilotti (1976) reported recovery in light reduction plots one to six months after removal of light reduction screens. Why eelgrass recovered in their experiments and not in the present study is not clear.

In addition to density of shoots of Z. marina, shoot biomass and chlorophyll content of the leaves were measured to indicate the effects of light reduction. Shoot biomass generally mirrored shoot density as an indicator of the effects of light reduction (Table 1). However, because shoot biomass is a destructive sampling technique, the limited sampling precludes valid statistical comparisons and provides no indication of delayed effects or of recovery. One of the disadvantages of shoot density as a measure of the effects of light reduction is that shoots that survive (or regenerate), no matter how small are counted equally with large healthy shoots. This contrast was especially evident at the subtidal plots where large shoots with leaves more than 1 m long were being counted equally with shoots with leaves about 0.2 m tall. The leaf dry weight per shoot in the light reduction plots vs. control plots at the end of the subtidal spring and summer experiments (Table 1) and at the end of the intertidal spring, summer, and autumn experiments (Table 3) illustrate this contrast. Thus, light reduction both reduced density and reduced the weight of the surviving shoots.

The concentration of chlorophyll and the chlorophyll a/b rations were not affected by the light reduction screens. This result contrasts with the increased chlorophyll content and increasing chlorophyll a/b ratios reported for Z. marina at increasing depths (Dennison and Alberte 1986) and for reduced light levels (Olesen and Sand-Jensen 1993).

In conclusion, this study has shown that short-term (8 - 12 weeks) light reduction to about 10% has a greater effect on density of Z. marina during spring and summer than it does in autumn and winter; that such light reduction may have delayed effects when light is reduced during autumn and winter; that intertidal Z. marina may be more sensitive to light reduction than subtidal Z. marina; and that recovery of Z. marina from even short-term light reduction may take more than one year.

LITERATURE CITED

Bach, S.D. and M.N. Josselyn. 1978. Mass blooms of the alga Cladophora in Bermuda. *Marine Pollution Bulletin*. 9: 34-37.

Backman, T.W. and D.C. Barilotti. 1976. Irradiance reduction: effects on standing crops of the eelgrass Zostera marina in a coastal lagoon. *Marine Biology*. 34: 33-40.

Baldwin, J.R. and J.R. Lovvorn. 1994. Habitats and tidal accessibility of the marine foods of dabbling ducks and brant in Boundary Bay, British Columbia. *Marine Biology*. 120: 627-638.

Bell, S.S., K. Walters and J.C. Kern. 1984. Meiofauna from seagrass habitats: a review and prospectus for future research. *Estuaries*. 7: 331-338.

Bulthuis, D.A. 1983a. Effects of in situ light reduction on density and growth of the seagrass Heterozostera tasmanica (Martens ex Aschers.) den Hartog in Western Port, Victoria, Australia. *Journal of Experimental Marine Biology and Ecology*. 67: 91-103.

Bulthuis, D.A. 1983b. Effects of temperature on the photosynthesis-irradiance curve of the Australian seagrass, Heterozostera tasmanica. *Marine Biology Letters*. 4: 47-57.

Bulthuis, D.A. 1987. Effects of temperature on photosynthesis and growth of seagrasses. *Aquatic Botany*. 27: 27-40.

Bulthuis, D.A. 1991. *Distribution of habitats and summer standing crop of seagrasses and macroalgae in Padilla Bay, Washington, 1989*. 35 pp. Washington State Dept. Ecology: Mount Vernon, Washington. Padilla Bay National Estuarine Research Reserve Technical Report No. 2.

Bulthuis, D.A. 1995. Distribution of seagrasses in a North Puget Sound Estuary: Padilla Bay, Washington, U.S.A. *Aquatic Botany*. 50: 99-105.

Bulthuis, D.A., G.W. Brand and M.C. Mobley. 1984. Suspended sediments and nutrients in water ebbing from seagrass-covered and denuded tidal mudflats in a southern Australian embayment. *Aquatic Botany*. 20: 257-266.

Bulthuis, D.A. and A.M. Conrad. 1995. *Guemes Channel and Padilla Bay: surface currents during flood tide*. 133 pp. Washington State Department of Ecology: Mount Vernon, Washington. Padilla Bay National Estuarine Research Reserve Technical Report No. 15.

Bulthuis, D.A. and R.A. Cowdell. 1982. *An annotated bibliography of eutrophic marine ecosystems*. 30 pp. Queenscliff, Victoria, Australia. Marine Science Laboratories Queenscliff Technical Report 19.

Bulthuis, D.A. and W.J. Woelkerling. 1983. Seasonal variation in standing crop, density and leaf growth rate of the seagrass, Heterozostera tasmanica (Martens ex Aschers.) den Hartog in Western Port and Port Phillip Bay, Victoria, Australia. *Aquatic Botany*. 16: 111-136.

Cambridge, M.L., A.W. Chiffings, C. Brittan, L. Moore and A.J. McComb. 1986. The loss of seagrass in Cockburn Sound, Western Australia. II. Possible causes of seagrass decline. *Aquatic Botany.* 2 4: 269-285.

Cambridge, M.L. and A.J. McComb. 1984. The loss of seagrasses in Cockburn Sound, Western Australia. I. The time course and magnitude of seagrass decline in relation to industrial development. *Aquatic Botany.* 20: 229-243.

Cassidy, P.M. and G.L. McKeen. 1 986. *Padilla Bay baseline water quality record.* 472 pp. [Final report to Marine & Estuarine Management Division, NOAA, Grant No. NA85AA-D-CZ046]. Shannon Point Marine Center, Western Washington University: Anacortes, Washington. Washington State Department of Ecology, Padilla Bay National Estuarine Research Reserve Reprint Series No. 2.

Congdon, R.A. and A.J. McComb. 1979. Productivity of Ruppia: seasonal changes and dependence on light in an Australian estuary. *Aquatic Botany.* 6: 121-132.

Dawes, C.J. and D.A. Tomasko. 1988. Depth distribution of Thalassia testudinum in two meadows on the west coast of Florida; a difference in effect of light availability. *Marine Ecology (Pubblicazioni della Stazione Zoologica di Napoli I).* 9: 123-130.

Den Hartog, C. 1994. Suffocation of a littoral Zostera bed by Enteromorpha radiata. *Aquatic Botany.* 47: 21-28.

Dennison, W.C. and R. Alberte. 1982. Photosythetic responses of Zostera marina L (eelgrass) to in situ manipulations of light intensity. *Oecologia (Berlin).* 55: 137-144.

Dennison, W.C. 1987. Effects of light on seagrass photosynthesis, growth and depth distribution. *Aquatic Botany.* 27: 15-26.

Dennison, W.C. and R.S. Alberte. 1985. Role of daily light period in the depth distribution of Zostera marina (eelgrass). *Marine Ecology Progress Series.* 25: 51-61.

Dennison, W.C. and R.S. Alberte. 1986. Photoadaptation and growth of Zostera marina L. (eelgrass) transplants along a depth gradient. *Journal of Experimental Marine Biology and Ecology.* 98: 265-282.

Dinnel, P.A., D.A. Armstrong and R.O. McMillan. 1993. Evidence for multiple recruitment-cohorts of Puget Sound Dungeness crab, Cancer magister. *Marine Biology.* 115: 53-63.

Giesen, W.B.J., M.M. van Katwijk and C. Den Hartog. 1990. Eelgrass condition and turbidity in the Dutch Wadden Sea. *Aquatic Botany.* 37: 71-85.

Harlin, M.M. and B. Thorne-Miller. 1981. Nutrient enrichment of seagrass beds in a Rhode Island coastal lagoon. *Marine Biology.* 65: 221-229.

Harlin, M.M., B. Thorne-Miller and J.C. Boothroyd. 1982. Seagrass-sediment dynamics of a flood-tidal delta in Rhode Island (U.S.A.). *Aquatic Botany.* 14: 127-138.

Kemp, W.M., W.R. Boynton, R.R. Twilley, J.C. Stevenson and J.C. Means. 1983. The decline of submerged vascular plants in Upper Chesapeake Bay: summary of results concerning possible causes. *Marine Technological Society Journal.* 17: 78-89.

Kentula, M.E. and C.D. McIntire. 1986. The autecology and production dynamics of eelgrass (Zostera marina L.) in Netarts Bay, Oregon. *Estuaries.* 9: 188-199.

Larkum, A.W.D. and R.J. West. 1983. Stability, depletion and restoration of seagrass beds. *Proc. Linn. Soc. N.S.W.* 106: 201-212.

Livingston, R.J. 1987. Historic trends of human impacts on seagrass meadows in Florida. *Proceed. symp. subtropical-tropical seagrasses of the southeastern United States Fla. Mar. Res. Publ.* 42: 139-151.

Marsh, J.A., W.C. Dennison and R.S. Alberte. 1986. Effects of temperature on photosynthesis and respiration in eelgrass (Zostera marina L.). *Journal of Experimental Marine Biology and Ecology.* 101: 257-267.

McMillan, R.O., D.A. Armstrong and P.A. Dinnel. 1995. Comparison of intertidal habitat use and growth rates of two northern Puget Sound cohorts of 0+ age Dungeness crab, Cancer magister. *Estuaries.* 18: 390-398.

Neinhuis, P.H. and B.H.H. DeBree. 1980. Production and growth dynamics of eelgrass (Zostera marina) in brackish Lake Grevelingen (The Netherlands). *Netherlands Journal of Sea Research.* 14: 102-118.

Neverauskas, V.P. 1988. Response of a Posidonia community to prolonged reduction in light. *Aquatic Botany.* 31: 361-366.

Olesen, B. and K. Sand-Jensen. 1993. Seasonal acclimatization of eelgrass Zostera marina growth to light. *Marine Ecology Progress Series.* 94: 91-99.

Orth, R.J. and K.A. Moore. 1983. Chesapeake Bay: an unprecedented decline in submerged aquatic vegetation. *Science.* 222: 51-53.

Orth, R.J. and K.A. Moore. 1988. Distribution of Zostera marina L. and Ruppia maritima L. sensu lato along depth gradients in the lower Chesapeake Bay, U.S.A. *Aquatic Botany.* 32: 291-305.

Perkins, E.J. and O.J. Abbott. 1972. Nutrient enrichment and sand flat fauna. *Marine Pollution Bulletin.* 3: 70-72.

Phillips, R.C. 1972. *Ecological life history of Zostera marina L. (eelgrass) in Puget Sound, Washington.* M.S. Thesis, University of Washington: Seattle, WA. 154 pp.

Phillips, R.C. 1984. *The ecology of eelgrass meadows in the Pacific Northwest: a community profile.* 85 pp. U.S. Fish Wildl. Serv.: FWS/OBS-84/24.

Phillips, R.C. 1980. Responses of transplanted and indigenous Thalassia testudinum Banks ex Konig and Halodule wrightii Aschers. to sediment loading and cold stress. *Contrib. Mar. Sci.* 23: 79-87.

Phillips, R.C. 1974. Transplantation of seagrasses, with special emphasis on eelgrass, Zostera marina L. *Aquaculture.* 4: 161-176.

Pollard, D.A. 1984. A review of ecological studies on seagrass-fish communities, with particular reference to recent studies in Australia. *Aquatic Botany.* 18: 3-42.

Powell, G.V.N. and F.C. Schaffner. 1991. Water trapping by seagrasses occupying bank habitats in Florida Bay. *Estuarine Coastal and Shelf Science.* 32: 43-60.

Sand-Jensen, K. 1975. Biomass, net production and growth dynamics in an eelgrass (Zostera marina L.) population in Vellerup Vig, Denmark. *Ophelia.* 14: 185-201.

Shepherd, S.A., A.J. McComb, D.A. Bulthuis, V. Neverauskas, D.A. Steffensen and R. West. 1989. Decline of seagrasses. *Biology of seagrasses, a treatise on the biology of seagrasses with special reference to the Australian region.* (editors. A.W.D. Larkum, A.J. McComb and S.A. Shepherd) pp. 346-393. Elsevier: Amsterdam.

Short, F.T. 1983. The response of interstitial ammonium in eelgrass (Zostera marina L.) beds to environmental perturbations. *Journal of Experimental Marine Biology and Ecology.* 68: 195-208.

Silberstein, K., A.W. Chiffings and A.J. McComb. 1986. The loss of seagrass in Cockburn Sound, Western Australia. III. The effect of epiphytes on productivity of Posidonia australis Hook. F. *Aquatic Botany.* 24: 355-371.

Simenstad, C.A., J.R. Cordell, R.C. Wissmar, K.L. Fresh, S.L. Schroder, M. Carr, G. Sanborn and M. Burg. 1988. *Assemblage structure, microhabitat distribution and food web linkages of epibenthic crustaceans in Padilla Bay National Estuarine Research Reserve, Washington.* 60 pp. Univ. Washington, Fisheries Research Institute: Seattle, WA. FRI-UW-8813. Padilla Bay National Estuarine Research Reserve Reprint Series No. 9, 1990.

Simenstad, C.A., J.R. Cordell, K.L. Fresh and M.I. Carr. 1995. *Trophic linkages from epibenthic crustaceans in littoral flat habitats: seasonal and regional comparisons.* 52 pp. University of Washington, School of Fisheries, Fisheries Research Institute: Seattle, Washington. FRI-UW-95__.

Stoner, A.W. and F.G. Lewis III. 1985. The influence of quantitative and qualitative aspects of habitat complexity in tropical seagrass meadows. *Journal of Experimental Biology and Ecology.* 94: 19-40.

Thom, R.M. 1980. Seasonality in low intertidal benthic marine algal communities in central Puget Sound, Washington, USA. *Botanica Marina.* 23: 7-11.

Thom, R.M. 1990. Spatial and temporal patterns in plant standing stock and primary production in a temperate seagrass system. *Botanica Marina.* 33: 497-510.

Thom, R.M. and R.G. Albright. 1990. Dynamics of benthic vegetation standing-stock, irradiance, and water properties in central Puget Sound. *Marine Biology.* 104: 129-141.

Thorne-Miller, B., M.M. Harlin, G.B. Thursby, M.M. Brady-Campbell and B.A. Dworetzky. 1983. Variations in the distribution and biomass of submerged macrophytes in five coastal lagoons in Rhode Island, U.S.A. *Botanica Marina.* 26: 231-242.

Tomasko, D.A. and C.J. Dawes. 1989. Evidence for physiological integration between shaded and unshaded short shoots of Thalassia testudinum. *Marine Ecology Progress Series.* 54: 299-305.

Webber, H.H., T.F.,. Mumford and J. Eby. 1987. *Remote sensing inventory of the seagrass meadow of the Padilla Bay National Estuarine Research Reserve. Areal extent and estimation of biomass.* 70 pp. Western Washington University, Huxley College of Environmental Studies: Bellingham, WA. Padilla Bay National Estuarine Research Reserve Reprint Series No. 6, 1990.

Williams, S.L. and M.H. Ruckelshaus. 1993. Effects of nitrogen availability and herbivory on eelgrass (Zostera marina) and epiphytes. *Ecology.* 74: 904-918.

Zieman, J.C. and R.G. Wetzel. 1980. Productivity in seagrasses: methods and rates. In: *Handbook of seagrass biology: an ecosystem perspective.* (editors. R.C. Phillips and C.P. McRoy) pp. 87-116. Garlands STPM: New York.

ACKNOWLEDGMENTS

Sincere thanks are due to many people who assisted me during this study, particularly to my all season diving partners Mark Olson and Sharon Riggs; and to Robin Cottrell who served as dive tender, cruise coordinator and data organizer and carried out all of the laboratory measurements and data entry. Thanks are also due to the Washington Conservation Crew, Mark Olson and Robin Cottrell for construction of the light reduction frames and screens; and to student interns Travis Shaw, Mary Jo Hartman, Brady Scott, Jennifer Crane, and Sherri Rodgers who assisted in the field work. This study was funded in part by a grant from the National Oceanic and Atmospheric Administration, National Ocean Service, Ocean and Coastal Resources Management, Sanctuaries and Reserves Division with funds appropriated for Section 315 of the Coastal Zone Management Act of 1972, as amended. The views expressed herein are those of the author and do not necessarily reflect the views of NOAA or any of its subagencies.

Table 1. Dry weight of leaves and roots/rhizomes of subtidal *Zostera marina* in control and experimental plots at which irradiance was reduced to 10% for about two months. Mean (± standard error of the mean) of four plots at start of experiment (2 sample quadrats per plot) and mean for one reduced irradiance plot and one control plot after treatment (3 or 4 sample quadrats per plot).

	Spring treatment leaf dry weight (g/0.0625 m2)		Summer treatment leaf dry weight (g/0.0625 m2)		Autumn treatment leaf dry weight (g/0.0625 m2)		Winter treatment leaf dry weight (g/0.0625 m2)	
	mean	s.e.	mean	s.e.	mean	s.e.	mean	s.e.
Leaf Dry Weight								
At start of experiment	4.14	±0.570	21.22	±3.587	6.27	±2.265	2.66	±0.387
Two months later								
Control	16.64		15.92		5.39		1.40	
Reduced irradiance	0.73		8.17		2.60		1.69	
Leaf Ash-free Dry Weight								
At start of experiment	2.74	±0.575	10.67	±1.742	3.03	±1.074	1.35	±0.208
Two months later								
Control	8.98		7.95		3.02		0.84	
Reduced irradiance	0.36		4.12		1.37		1.04	
Root/Rhizome Dry Weight								
At start of experiment	3.73	±1.100						
Two months later								
Control	6.54							
Reduced irradiance	0.73							
Root/Rhizome Ash-free Dry Weight								
At start of experiment	2.33	±0.725						
Two months later								
Control	4.84							
Reduced irradiance	0.40							
Leaf Dry Weight per Shoot								
	(g per shoot)							
At start of experiment	0.81	±0.381	2.26	±0.452	1.41	±0.392	0.83	±0.121
Two months later								
Control	1.86		1.63		0.88		0.34	
Reduced irradiance	0.87		0.96		0.62		0.37	

Table 2. Chlorophyll a & b in leaves of subtidal Zostera marina in experimental plots at whcih irradiance was reduced to 10% of control plots for ten to twelve weeks. Mean (± s.e.) of 3 to 4 plots (2 to 4 samples per plot).

		Chlorophyll a (µg chl/ sq cm)		Chlorophyll b (µg chl/ sq cm)		Chlorophyll a+b (µg chl/ sq cm)		Chlorophyll a/b	
		mean	±s.e.	mean	±s.e.	mean	±s.e.	mean	±s.e.
Spring Experiment									
Before treatment	(25 Mar 93)	20.4	±1.42	7.9	±0.59	28.3	±2.01	2.6	±0.02
After ten weeks	(2 Jun 93)								
Control		18.4	±1.65	7.2	±0.52	25.6	±2.16	2.5	±0.06
Treatment		20.6	±1.29	8.3	±0.51	28.9	±1.80	2.5	±0.01
Summer Experiment									
Before treatment	(25 Jun 93)	21.1	±2.12	8.7	±0.88	29.8	±3.00	2.4	±0.01
After five weeks	(27 Jul 93)								
Control		18.0	±1.24	7.3	±0.47	25.3	±1.70	2.5	±0.03
Treatment		17.2	±5.21	6.6	±2.15	23.8	±7.36	2.6	±0.06
After nine weeks	(26 Aug 93)								
Control		17.1	±0.26	7.2	±0.12	24.3	±0.39	2.4	±0.01
Treatment		12.7	±2.01	5.4	±0.87	18.0	±2.89	2.4	±0.01
Autumn Experiment									
Before treatment	(23 Sep 93)	19.5	±0.84	8.0	±0.35	27.4	±1.18	2.4	±0.02
After four weeks	(19 Oct 93)								
Control		23.3	±2.79	9.7	±1.12	32.9	±3.90	2.4	±0.03
Treatment		15.2	±0.86	6.4	±0.30	21.6	±1.13	2.4	±0.07
After twelve weeks	(14 Dec 93)								
Control		15.5	±1.33	5.9	±0.58	21.5	±1.91	2.6	±0.05
Treatment		14.7	±1.81	5.4	±0.78	20.1	±2.58	2.8	±0.08
Winter Experiment									
Before treatment	(21 Dec 93)	17.2	±1.02	6.7	±0.49	23.9	±1.51	2.6	±0.05
After five weeks	(26 Jan 94)								
Control		21.8	±0.56	8.7	±0.40	30.5	±0.94	2.5	±0.06
Treatment		20.6	±4.71	8.2	±1.88	28.8	±6.58	2.5	±0.06
After ten weeks	(3 Mar 94)								
Control		24.1	±3.64	9.4	±1.58	33.5	±5.22	2.6	±0.04
Treatment		18.6	±2.49	7.1	±1.04	25.8	±3.53	2.6	±0.05

Table 3. Dry weight of leaves and roots/rhizomes of intertidal *Zostera marina* in control and experimental plots at which irradiance was reduced to 10% for about two months. Mean (± standard error of the mean) of four plots at start of experiment (2 sample quadrats per plot) and mean for one reduced irradiance plot and one control plot after treatment (3 or 4 sample quadrats per plot).

Treatment	Spring treatment leaf dry weight (g/0.0625 m2)	Summer treatment leaf dry weight (g/0.0625 m2)		Autumn treatment leaf dry weight (g/0.0625 m2)		Winter treatment leaf dry weight (g/0.0625 m2)	
	mean	mean	s.e.	mean	s.e.	mean	s.e.
Leaf Dry Weight							
At start of experiment		11.34	±2.47	7.17	±1.21	2.64	±0.73
Two months later							
Control	7.87	6.47		4.51		3.81	
Reduced irradiance	0.09	1.44		0.75		2.80	
Leaf Ash-free Dry Weight							
At start of experiment		6.31	±1.28	3.47	±0.59	1.64	±0.43
Two months later							
Control	4.62	3.44		2.65		2.24	
Reduced irradiance	0.05	0.68		0.46		1.67	
Root/Rhizome Dry Weight							
At start of experiment		3.58	±0.73	3.86	±0.64		
Two months later							
Control	5.07						
Reduced irradiance	0.72						
Root/Rhizome Ash-free Dry Weight							
At start of experiment		2.24	±0.65	2.63	±0.46		
Two months later							
Control	2.97						
Reduced irradiance	0.34						
Leaf Dry Weight per Shoot							
	(g per shoot)						
At start of experiment		0.51	±0.049	0.47	±0.080	0.14	±0.024
Two months later							
Control	0.41	0.44		0.24		0.11	
Reduced irradiance	0.27	0.18		0.08		0.10	

Table 4. Chlorophyll a & b in leaves of intertidal Zostera marina in experimental plots at whcih irradiance was reduced to 10% of control plots for nine to eleven weeks. Mean (± s.e.) of 3 to 4 plots (2 to 4 samples per plot).

		Chlorophyll a (µg chl/ sq cm)		Chlorophyll b (µg chl/ sq cm)		Chlorophyll a+b (µg chl/ sq cm)		Chlorophyll a/b	
		mean	±s.e.	mean	±s.e.	mean	±s.e.	mean	±s.e.
Spring Experiment									
Before treatment	(26 Mar 93)	16.2	±0.78	5.7	±0.27	22.0	±1.04	2.8	±0.03
After nine weeks	(26 May 93)								
Control		15.7	±0.45	5.8	±0.08	21.5	±0.41	2.7	±0.09
Treatment		14.7		5.8		20.5		2.5	
Summer Experiment									
Before treatment	(22 Jun 93)	15.4	±1.13	5.3	±0.31	20.7	±1.43	2.9	±0.07
After four weeks	(20 Jul 93)								
Control		13.4	±1.82	5.2	±0.76	18.6	±2.57	2.6	±0.08
Treatment		18	±2.39	7.1	±1.05	25.1	±3.42	2.6	±0.09
After nine weeks	(27 Aug 93)								
Control		14.2	±1.20	5.7	±0.49	19.9	±1.67	2.5	±0.06
Treatment		15.7	±2.76	7.1	±1.13	22.8	±3.88	2.2	±0.08
Autumn Experiment									
Before treatment	(15 Sep 93)	14.4	±0.88	5.1	±0.56	19.5	±1.25	3.2	±0.64
After four weeks	(14 Oct 93)								
Control		13.1	±1.85	4.8	±0.69	17.9	±2.55	2.7	±0.02
Treatment		18.8	±2.25	7.2	±0.66	26.0	±2.90	2.6	±0.09
After nine weeks	(16 Nov 93)								
Control		20.7	±5.51	7.3	±1.80	28.0	±7.30	2.8	±0.07
Treatment		29.5	±3.61	11.1	±1.26	40.6	±4.86	2.7	±0.06
Winter Experiment									
Before treatment	(22 Dec 93)	24.9	±1.39	8.7	±0.44	33.6	±1.82	2.8	±0.03
After four weeks	(21 Jan 94)								
Control		19.1	±2.92	7.2	±1.16	26.3	±4.08	2.7	±0.04
Treatment		20.3	±1.76	7.8	±0.71	28.1	±2.46	2.6	±0.03
After eleven weeks	(8 Mar 94)								
Control		21.1	±0.95	7.2	±0.33	28.3	±1.18	2.9	±0.12
Treatment		17.4	±1.15	5.8	±0.22	23.2	±1.36	3.0	±0.09

Table 5. Summary of effects of nine to twelve weeks of light reduction to 10% on density of Zostera marina during four seasons at a subtidal and intertidal site in Padilla Bay, Washington. Potential recovery was monitored for 19 months (Spring '93) to 7 months (Spring '94) after removal of the light reduction screens. When there was no effect, possible recovery is not applicable (NA).

	Immediate effect?	**Delayed effect?**	**Recovery?**
Subtidal			
Spring '93	Yes	Yes	No
Summer '93	Yes	Yes?	No
Autumn '93	No	No	NA
Winter '93	No	Yes	Yes/No
Spring '94	Yes	No	No
Intertidal			
Spring '93	Yes	No	No
Summer '93	Yes	No	Yes/No
Autumn '93	No	Yes	Yes/No
Winter '93	No	No	NA
Spring '94	Yes	No	No

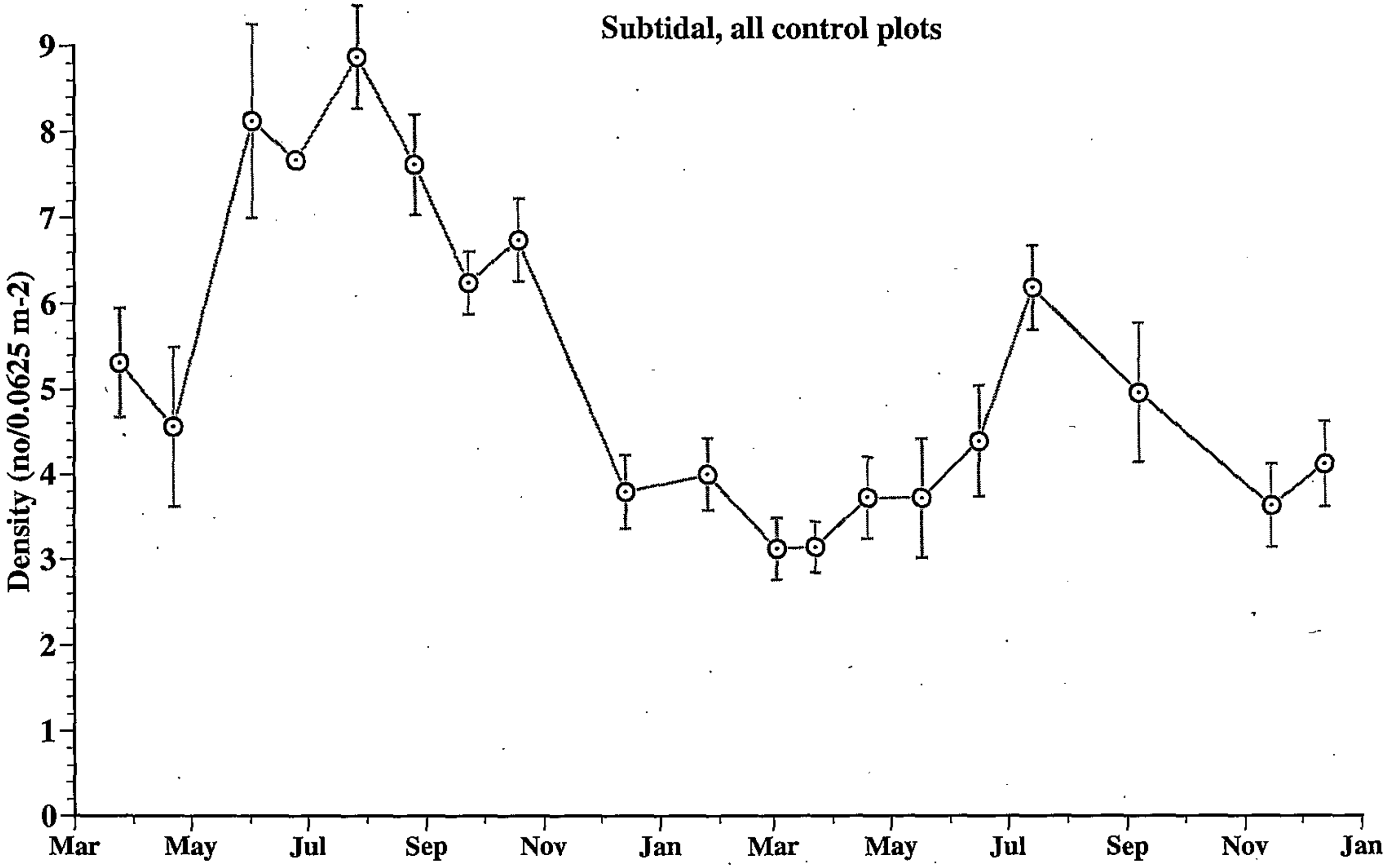


Figure 2. Density of Zostera marina in control plots at the subtidal site for the duration of the experiment (March 1993 to December 1994); mean ± s.e. (n=3 plots in March 1993 increasing up to 12 plots by March 1994 and thereafter).

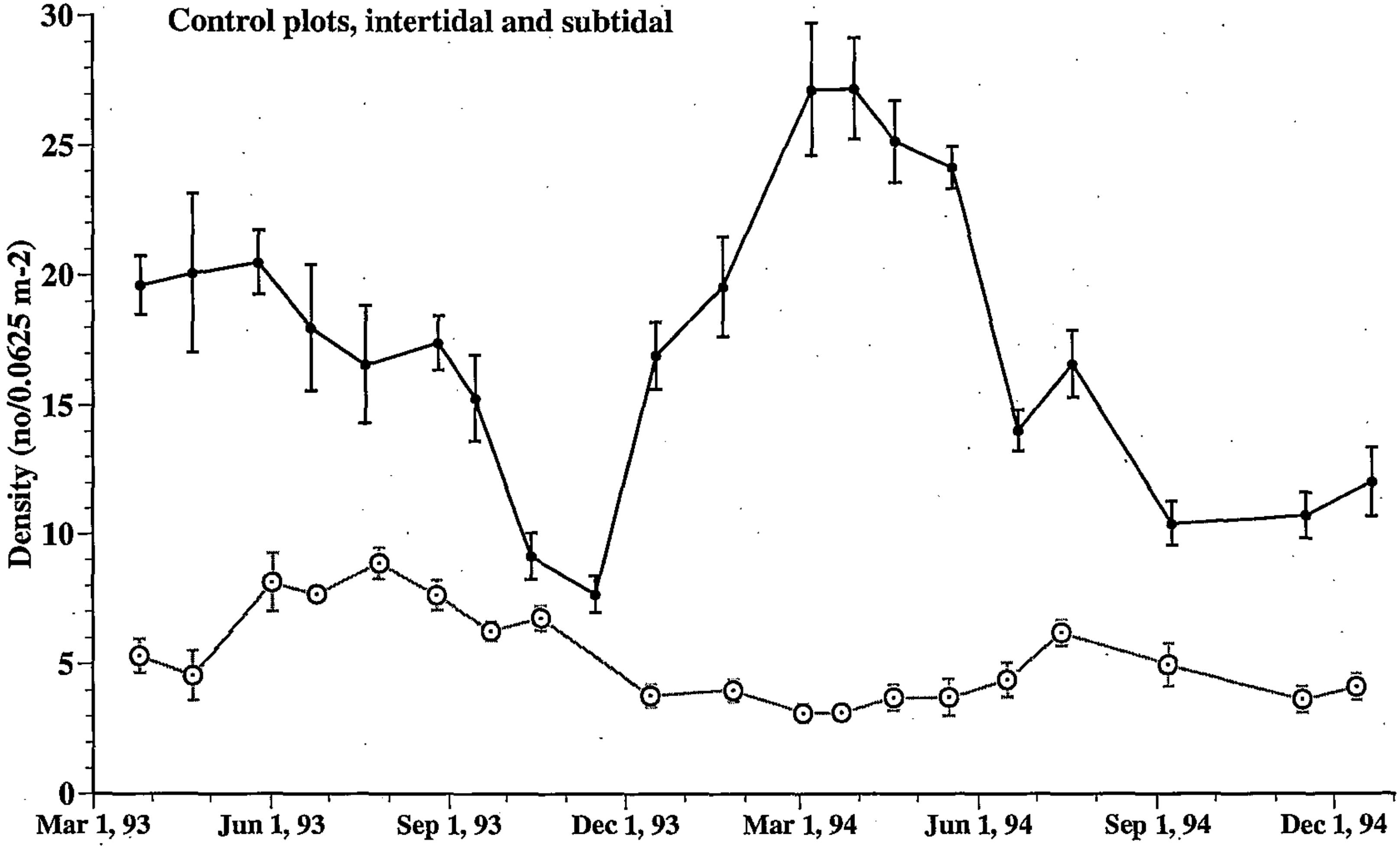


Figure 3. Density of Zostera marina in control plots at the intertidal site (closed circles) and the subtidal site (open circles) from March 1993 to December 1994. Mean ± s.e., n=3 plots in March 1993 increasing up to 12 plots by March 1994 and thereafter.

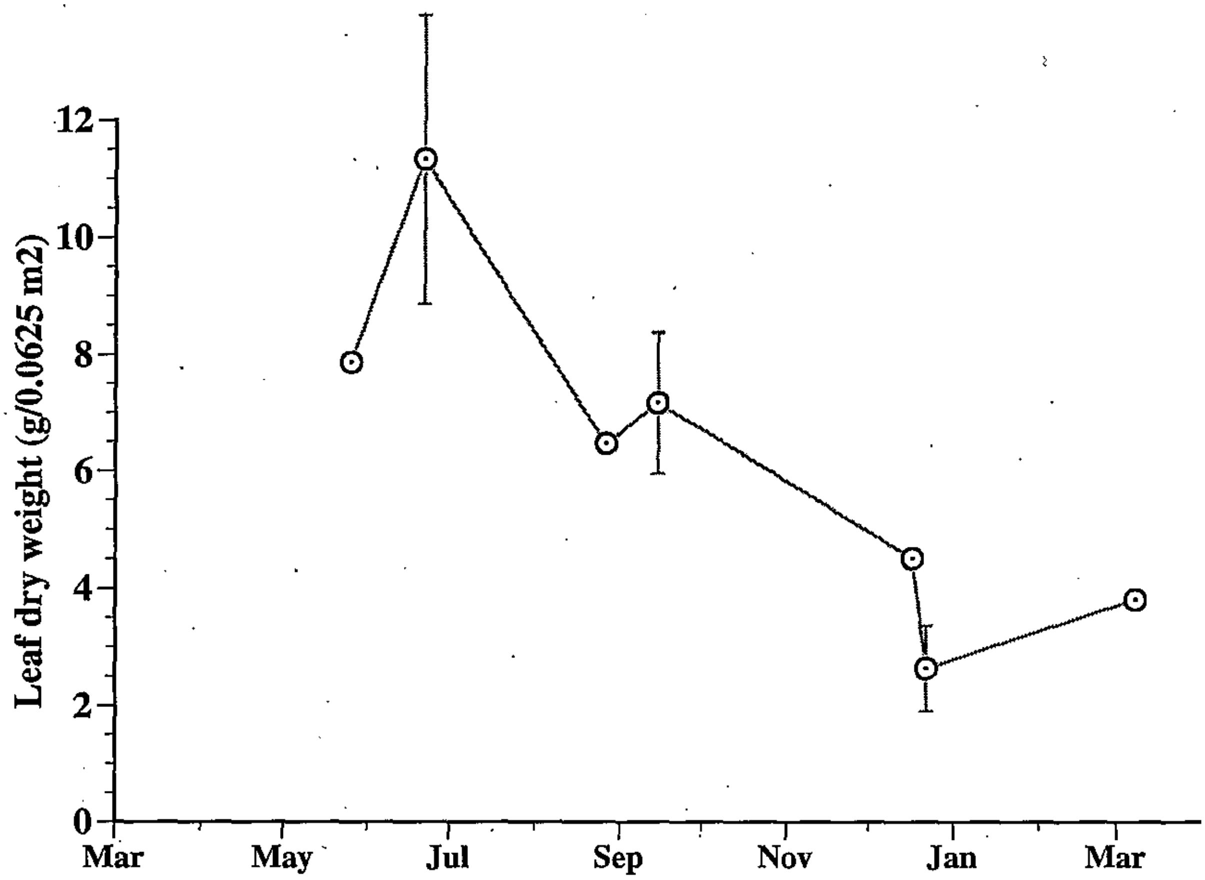


Figure 4. Leaf dry weight of Zostera marina in the control plots at the intertidal experimental site from May 1993 to March 1994. Mean ± s.e., n=1 to 3 plots.

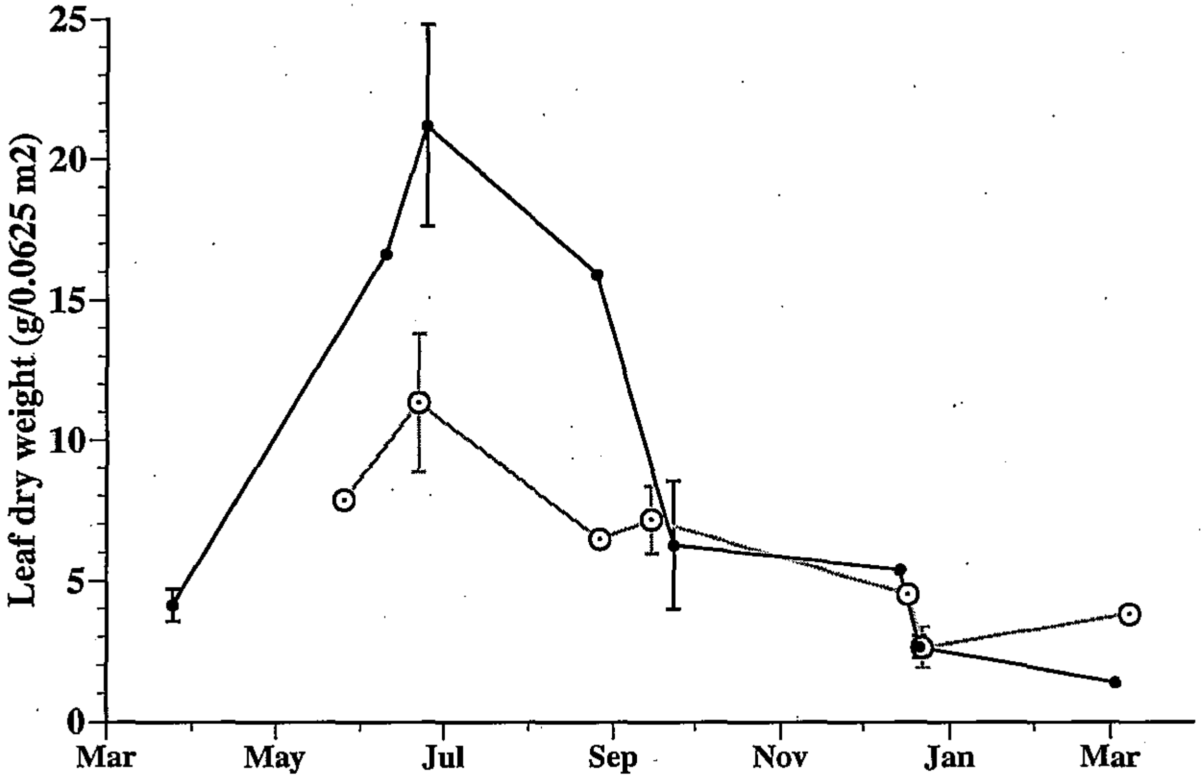


Figure 5. Leaf dry weight of Zostera marina in the control plots at the intertidal site (open circles) and at the subtidal site (closed circles) from May 1993 to March 1994. Mean ± s.e., n=1 to 3 plots.

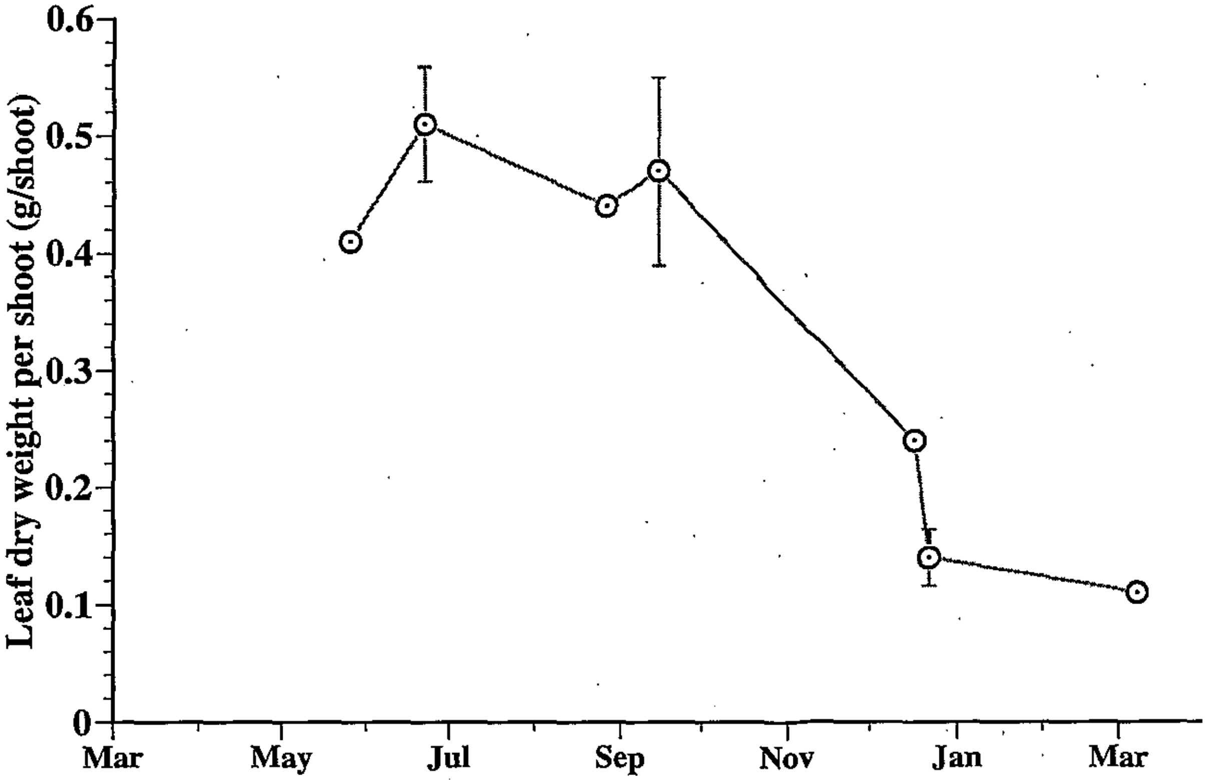


Figure 6. Leaf dry weight per shoot of Zostera marina in the control plots at the intertidal site from May 1993 to March 1994. Mean ± s.e., n=1 to 3 plots.

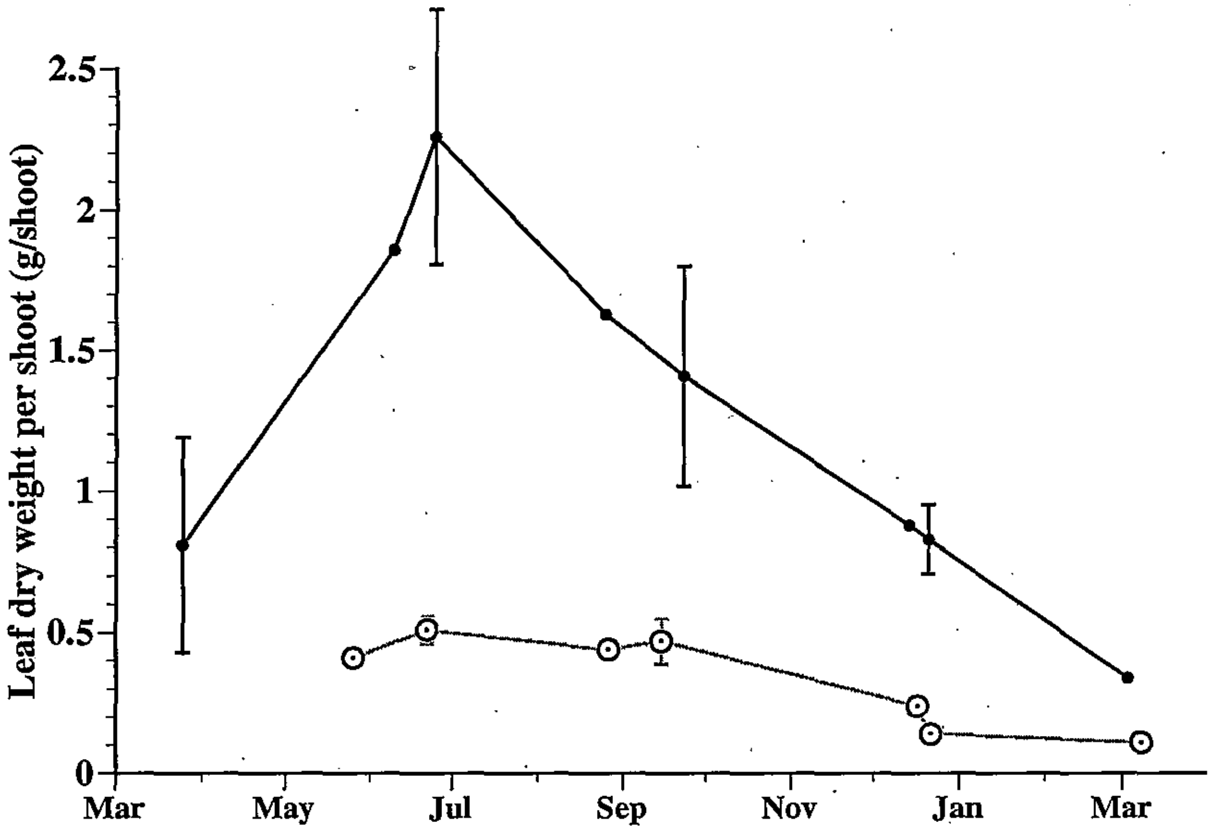


Figure 7. Leaf dry weight per shoot of Zostera marina in the control plots at the intertidal site (open circles) and at the subtidal site (closed circles) from May 1993 to March 1994. Mean ± s.e., n=1 to 3 plots.

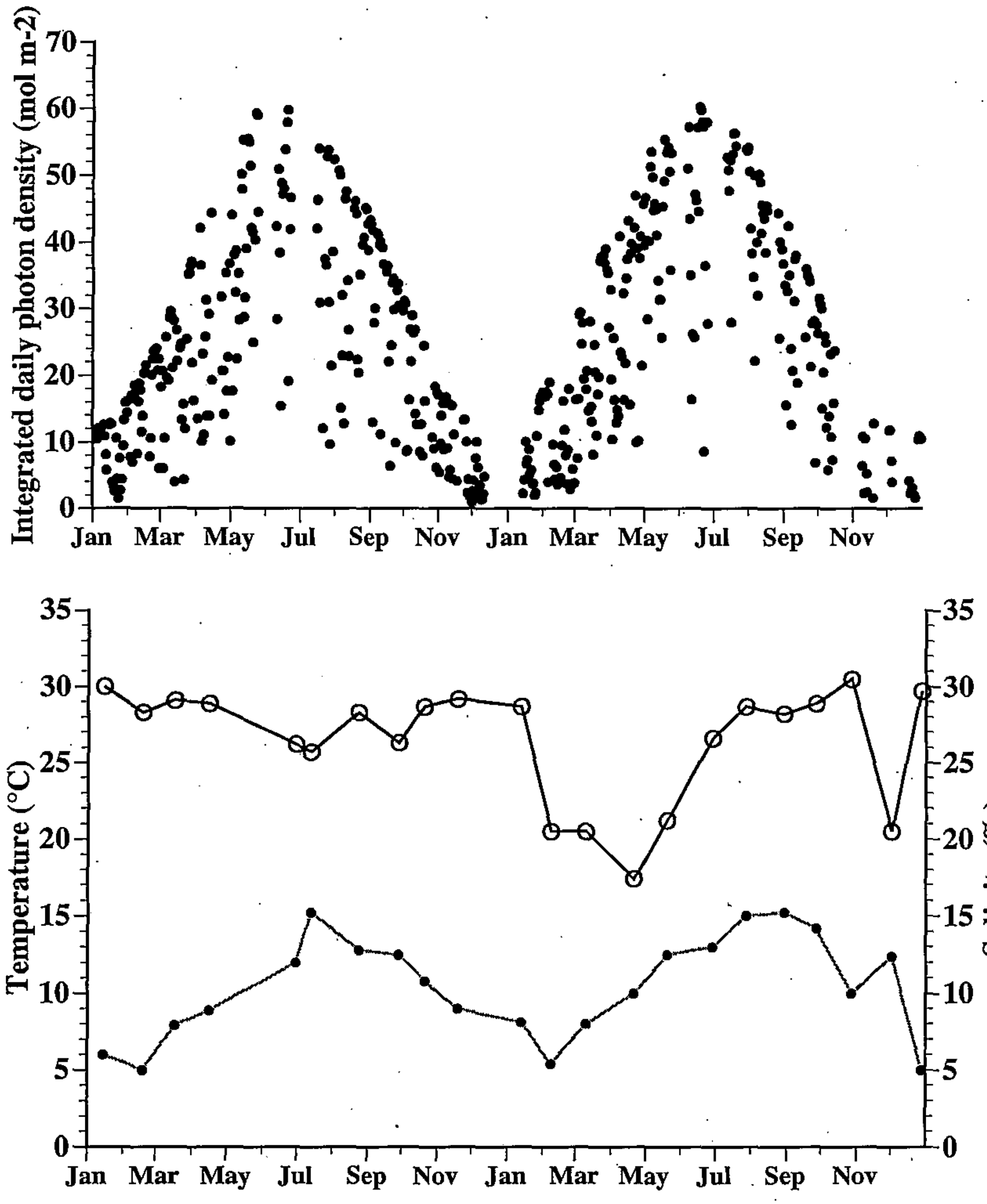


Figure 8. Integrated daily photon density (photosynthetically active radiation, 400-700 nm) at Padilla Bay National Estuarine Research Reserve and monthly temperature (•) and salinity (O) measured at a mid-bay station during 1993 and 1994. [Light data are missing for days when the instrument was used for other experiments, in particular from 12 Dec 93 to 15 Jan 94.]

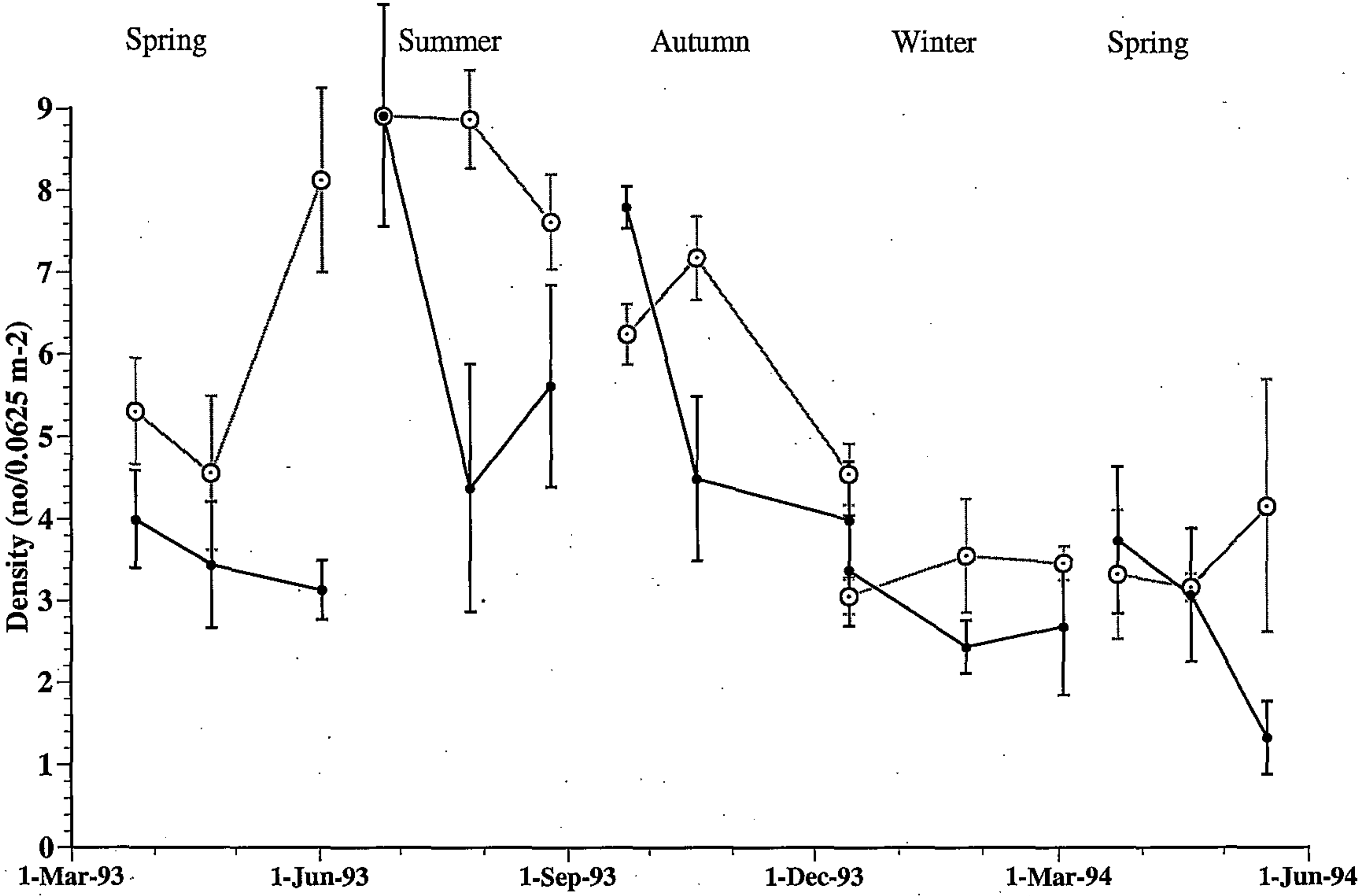


Figure 9. Density of Zostera marina in shaded and control plots at the subtidal site, mean ± s.e., n=3 plots. Shade experiments were initiated 3 months apart beginning in March 1993.

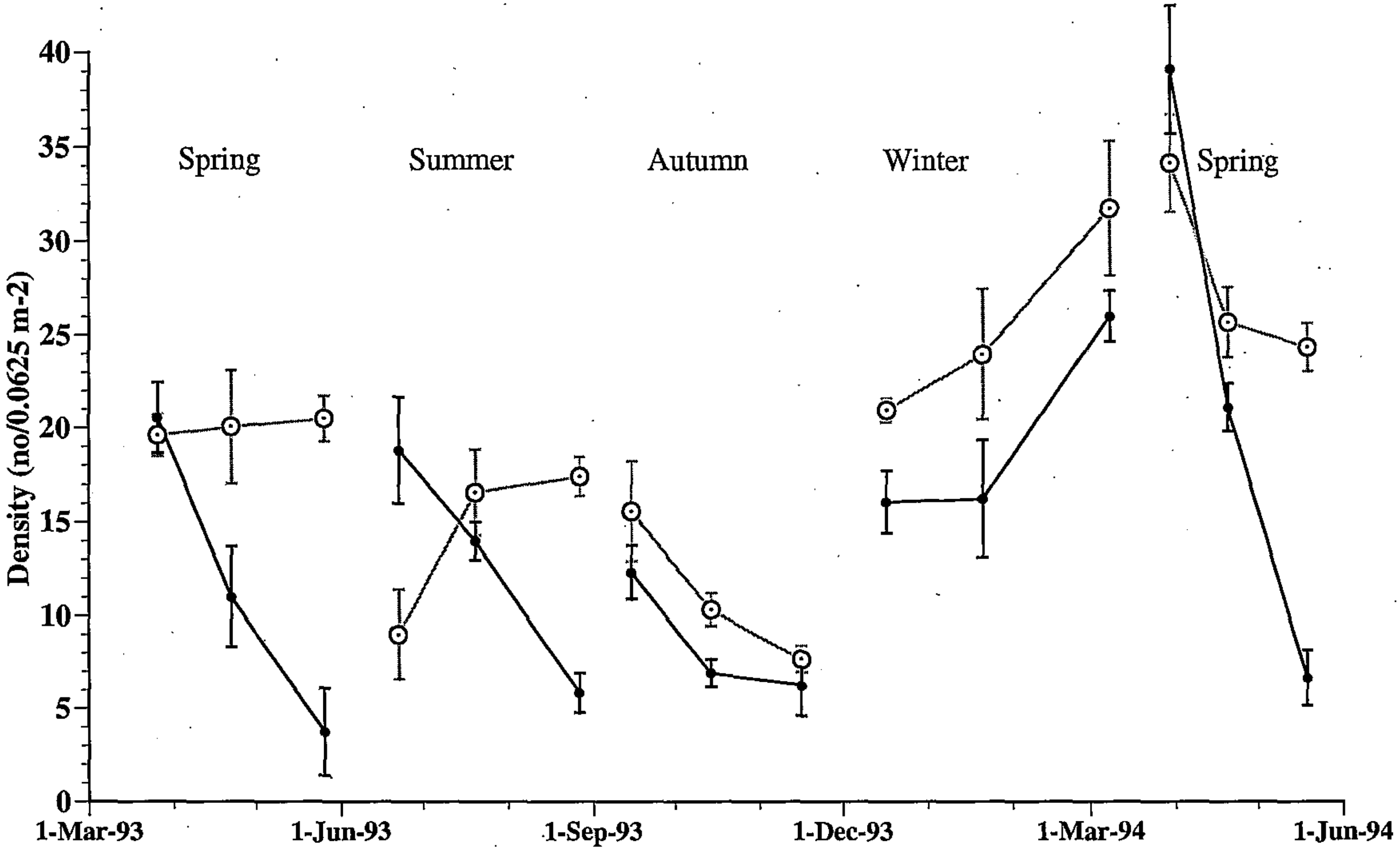


Figure 10. Density of Zostera marina in shaded and control plots at the intertidal site, mean ± s.e., n=3 plots. Shade experiments were initiated 3 months apart beginning in March 1993.

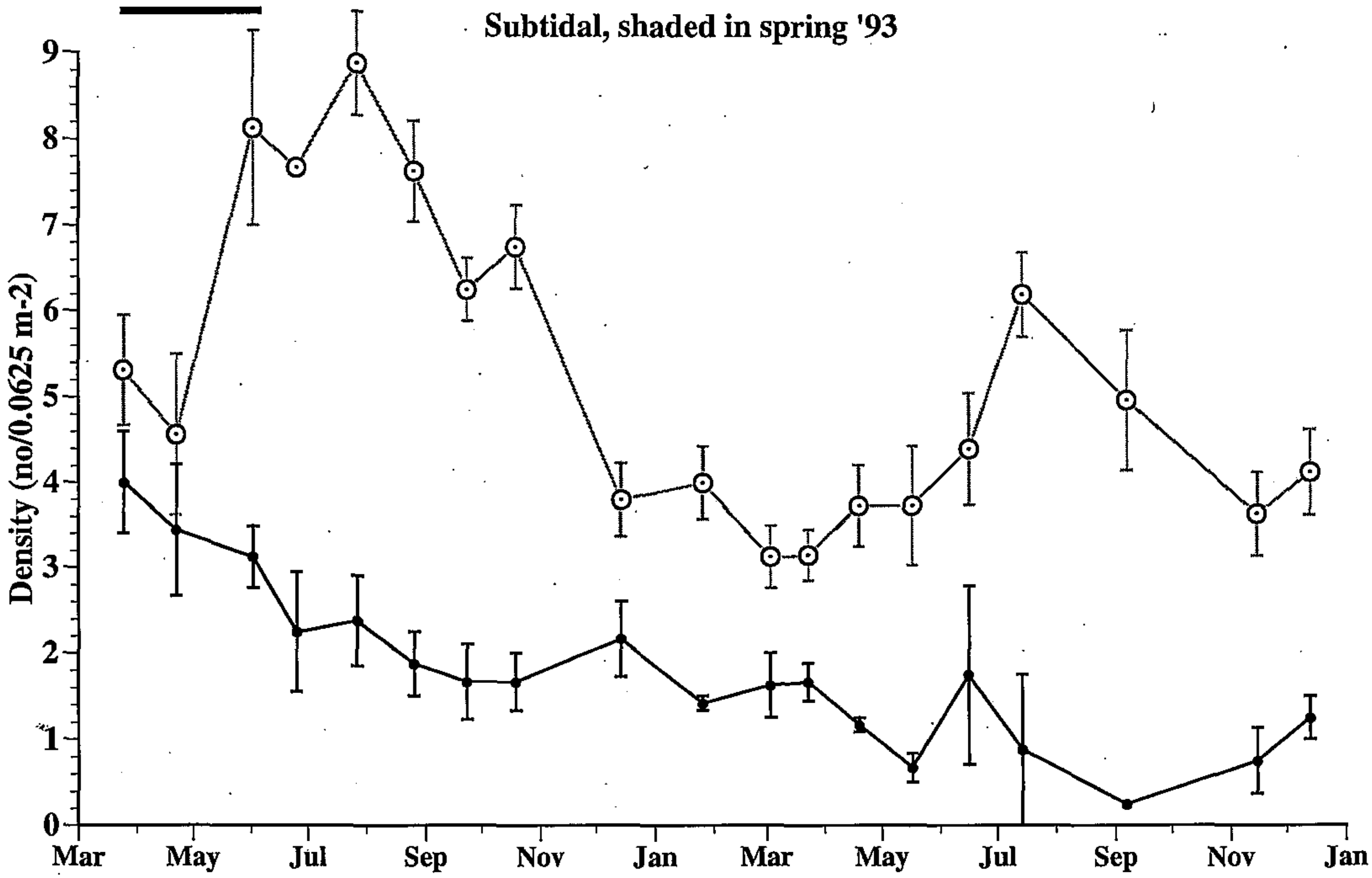


Figure 11. Density of Zostera marina at the subtidal site in plots (n=3) shaded for 10 weeks during spring 1993 (solid bar) and in control plots (n=3 to 12) from all experiments; mean ± s.e.

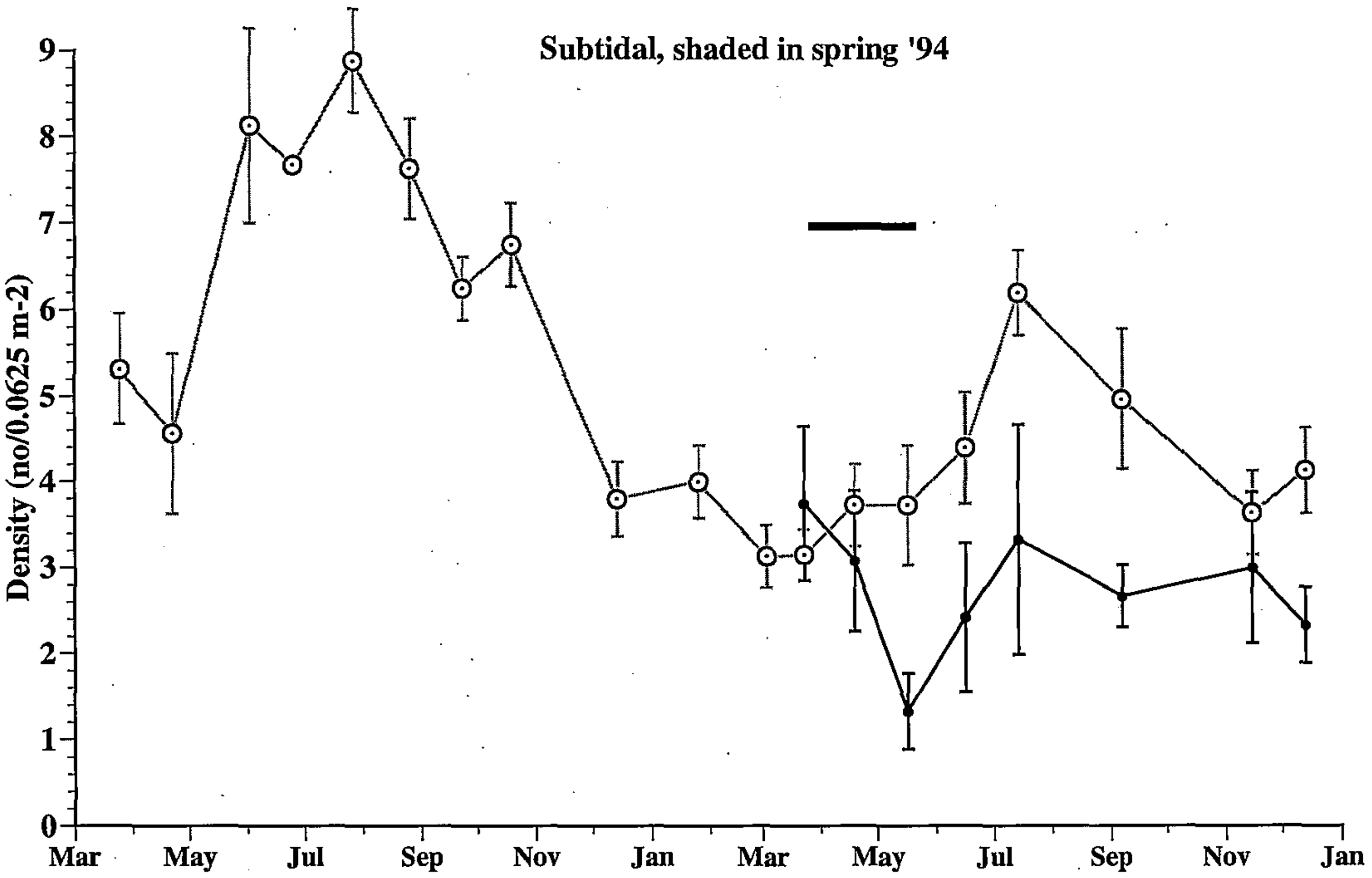


Figure 12. Density of Zostera marina at the subtidal site during 1993 and 1994 in plots (n=3) shaded for 8 weeks during spring 1994 (solid bar) and in control plots (n=3 to 12) from all experiments; mean ± s.e.

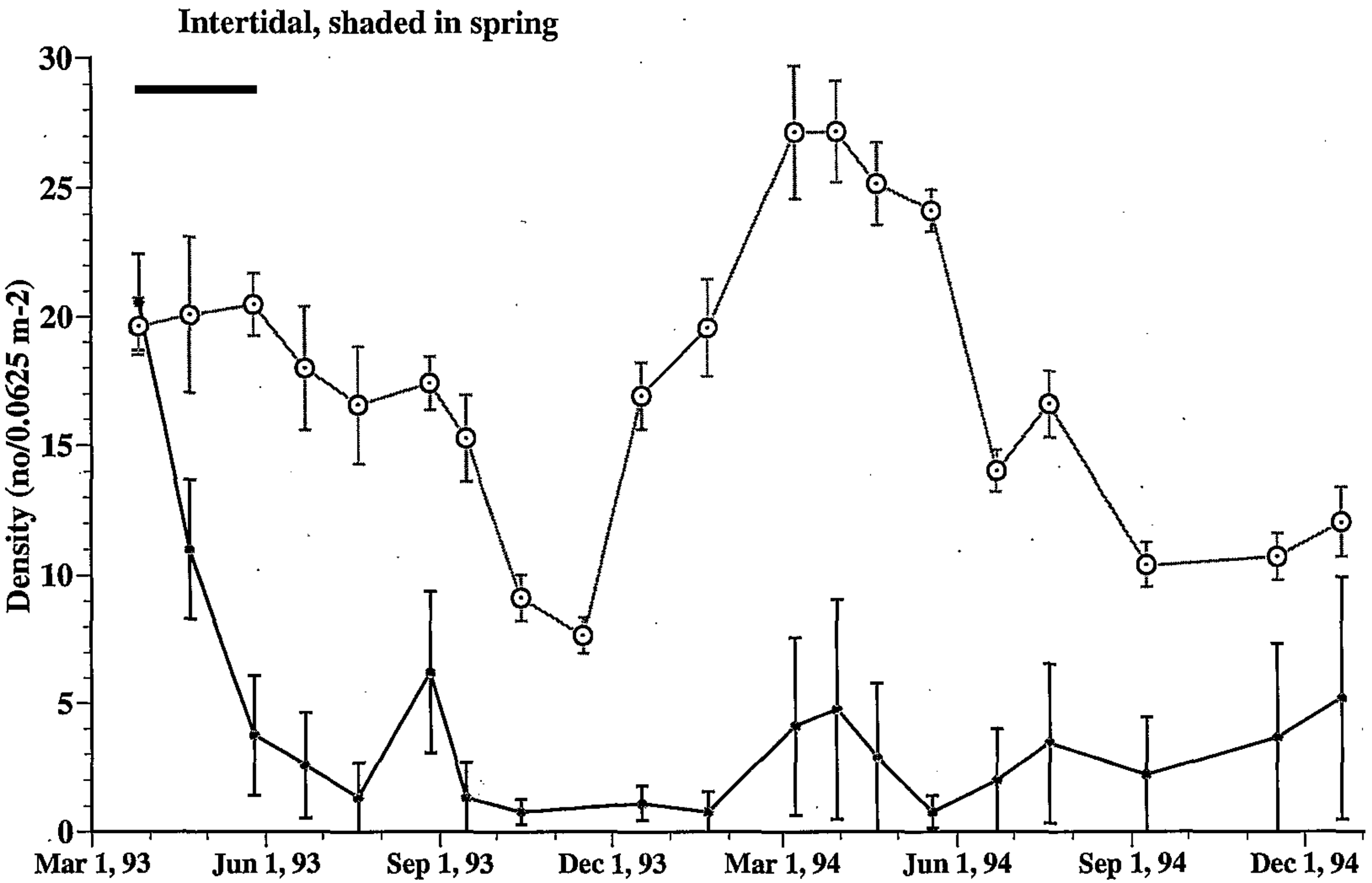


Figure 13. Density of Zostera marina at the intertidal site during 1993 and 1994 in plots (n=3) shaded for 9 weeks during spring 1993 (solid bar) and in control plots (n=3 to 12) from all experiments; mean ± s.e.

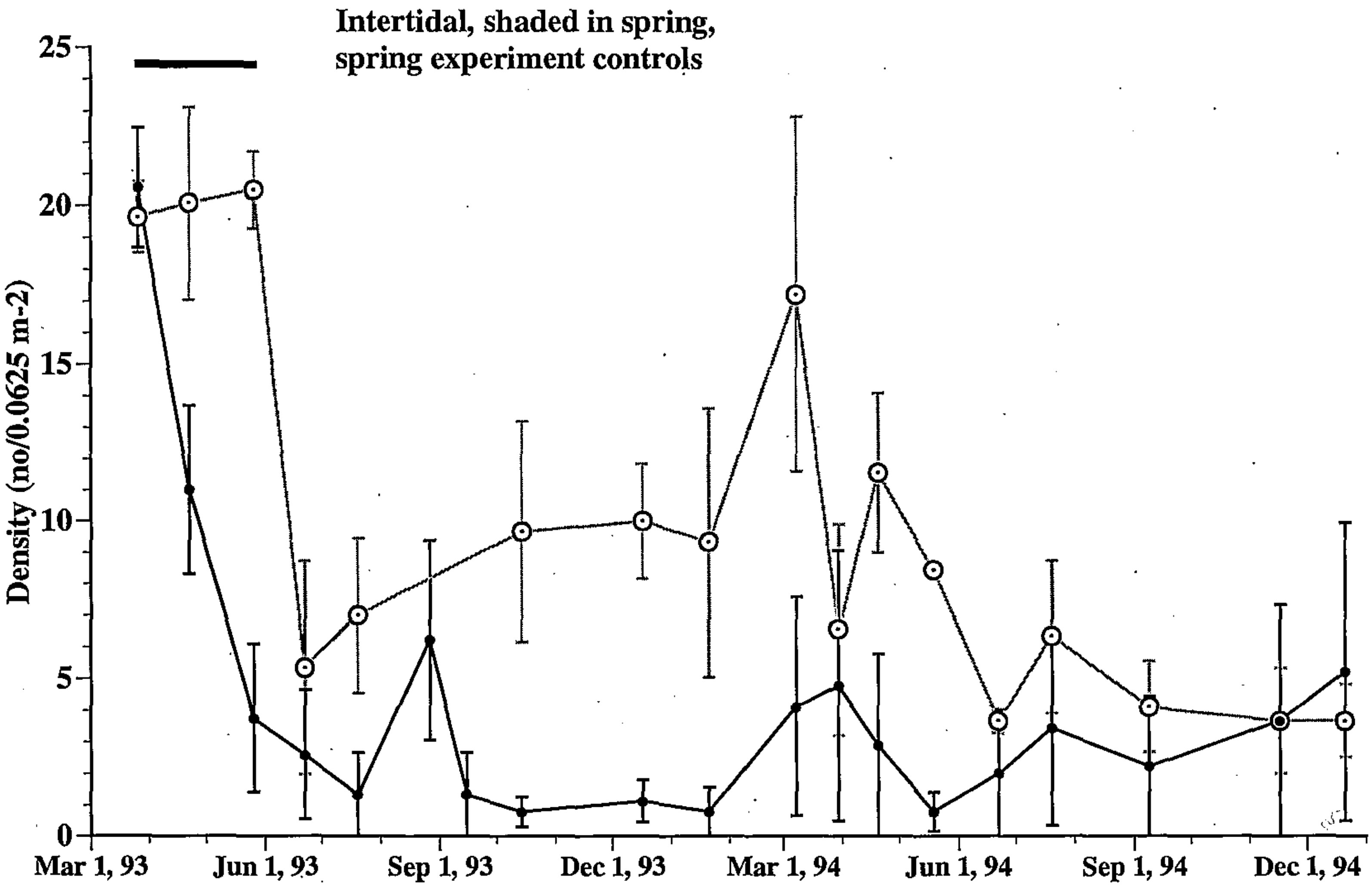


Figure 14. Density of Zostera marina at the intertidal site during 1993 and 1994 in plots (n=3) shaded for 9 weeks during spring 1993 (solid bar) and in control plots (n=3) from the spring experiment; mean ± s.e.

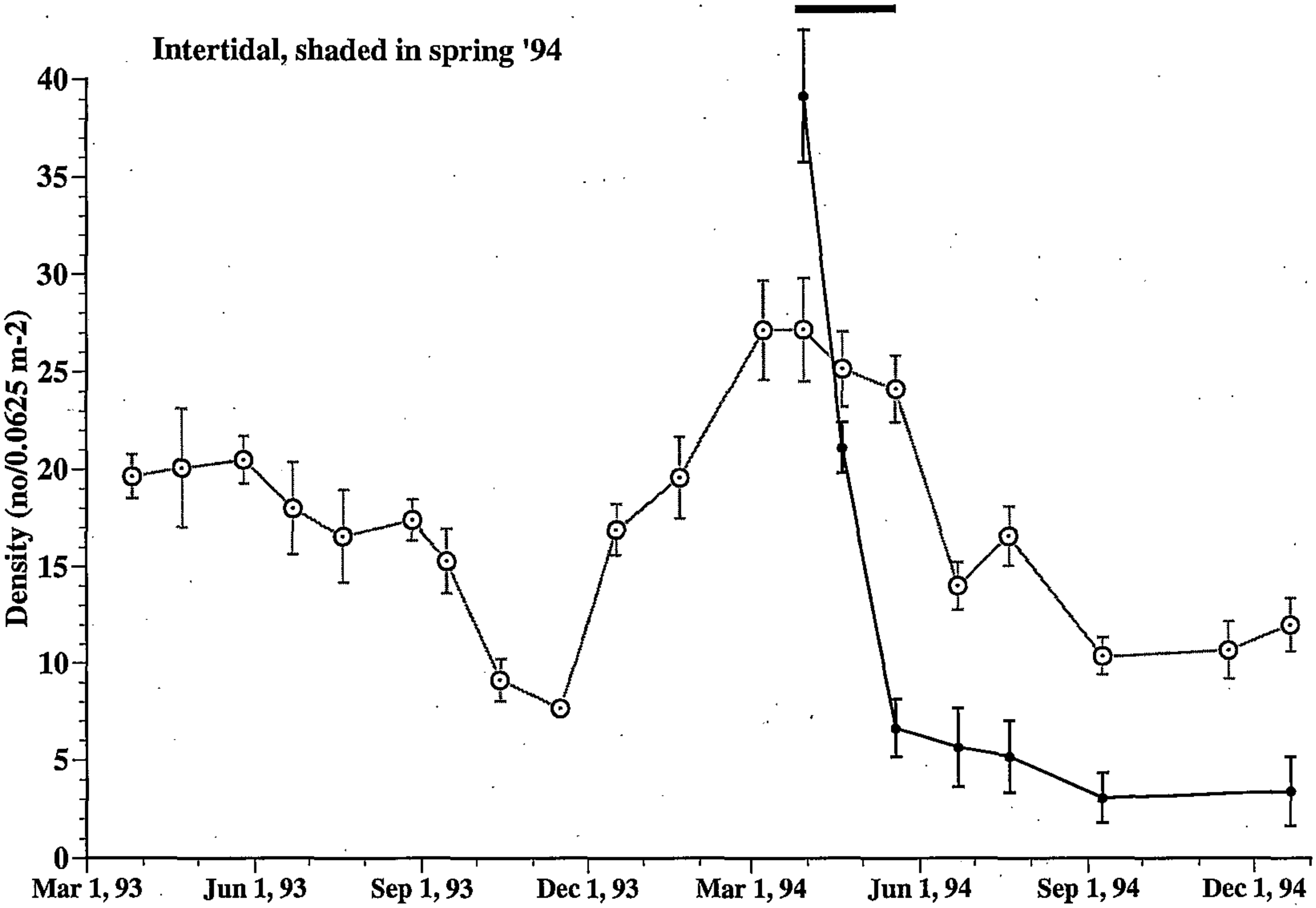


Figure 15. Density of <u>Zostera</u> <u>marina</u> at the intertidal site during 1993 and 1994 in plots (n=3) shaded for 7 weeks during spring 1994 (solid bar) and in control plots (n=3 to 12) from all experiments; mean ± s.e.

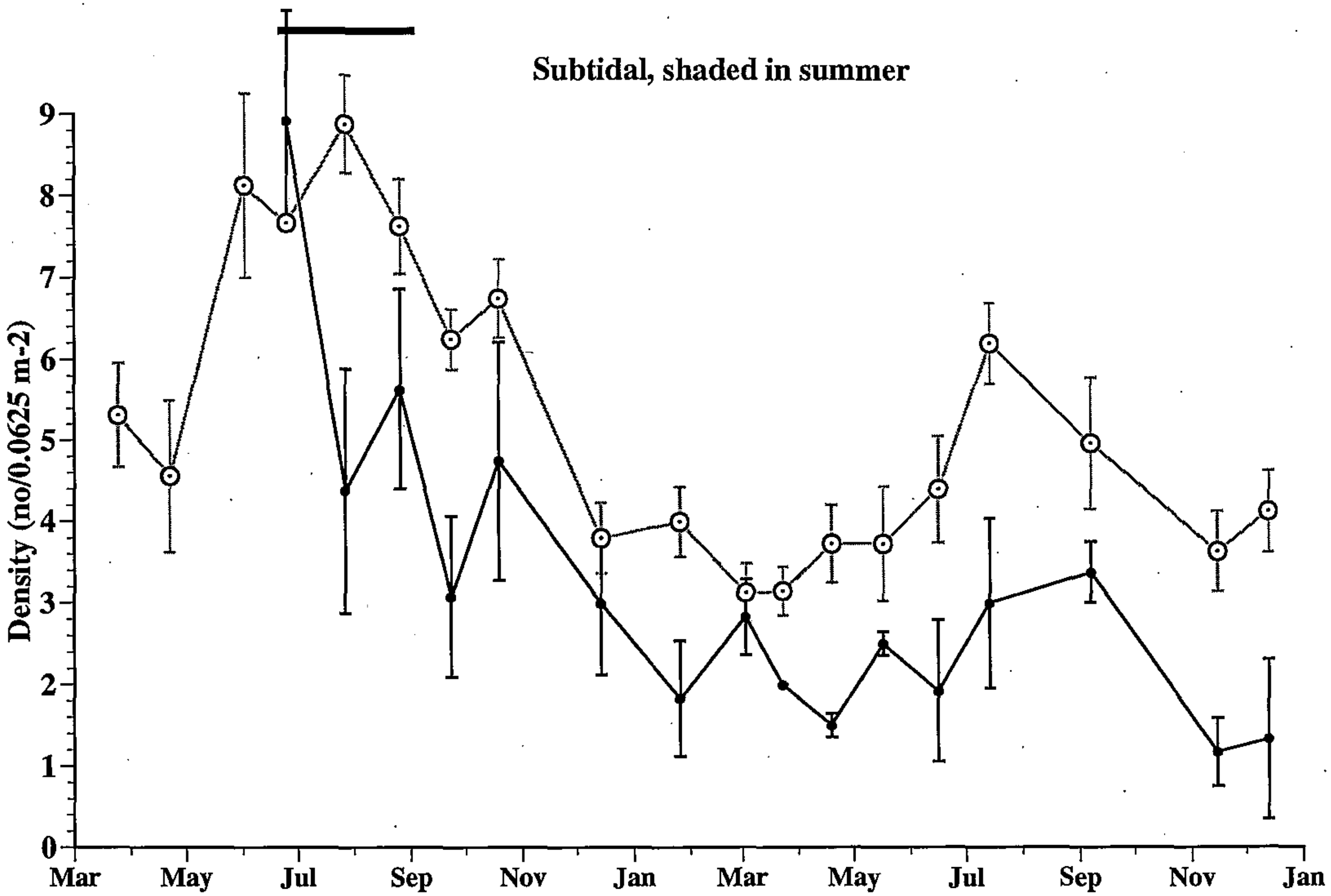


Figure 16. Density of Zostera marina at the subtidal site during 1993 and 1994 in plots (n=3) shaded for 9 weeks during summer (solid bar) and in control plots (n=3 to 12) from all experiments; mean ± s.e.

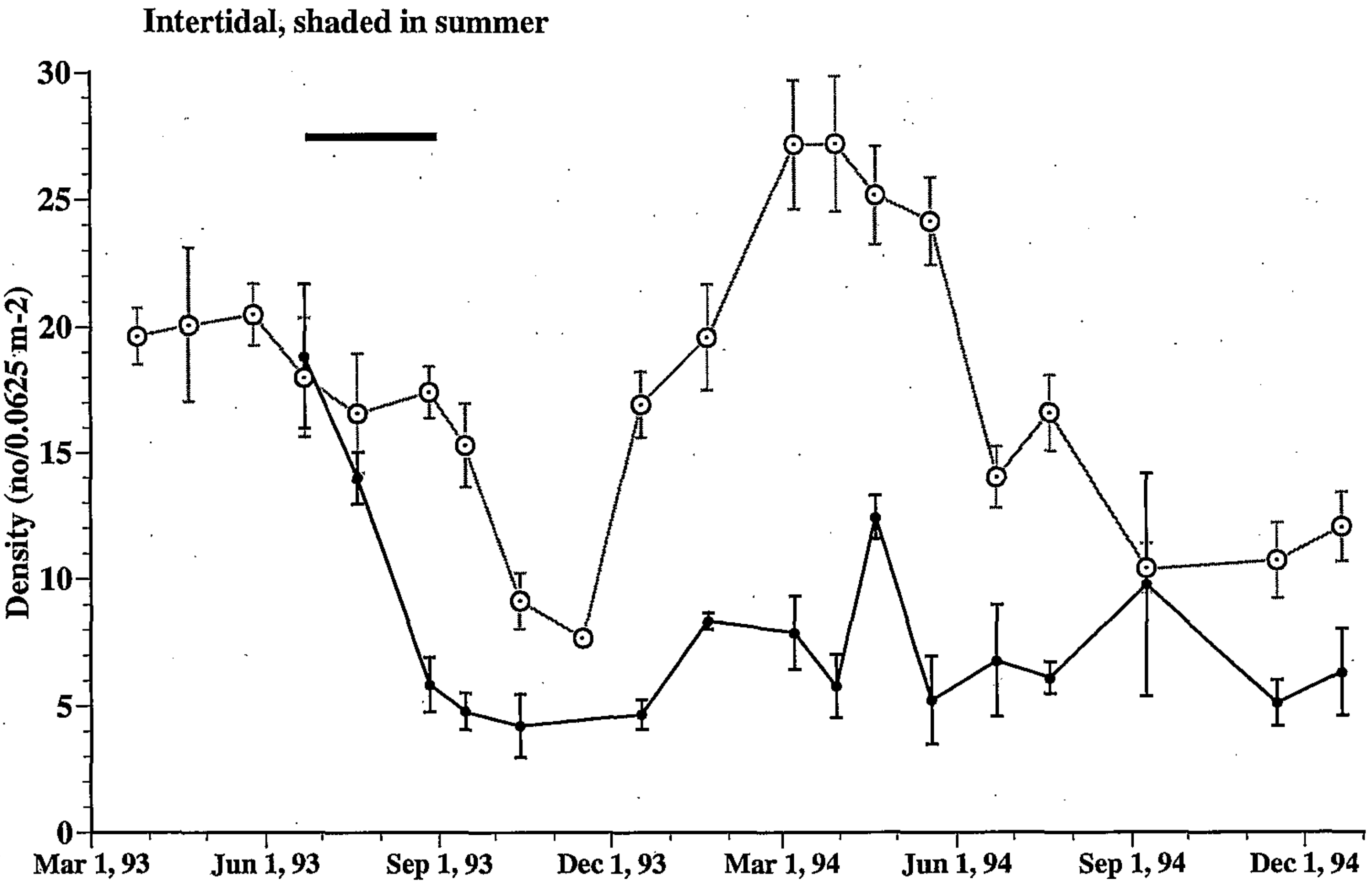


Figure 17. Density of Zostera marina at the intertidal site during 1993 and 1994 in plots (n=3) shaded for 9 weeks during summer (solid bar) and in control plots (n=3 to 12) from all experiments; mean ± s.e.

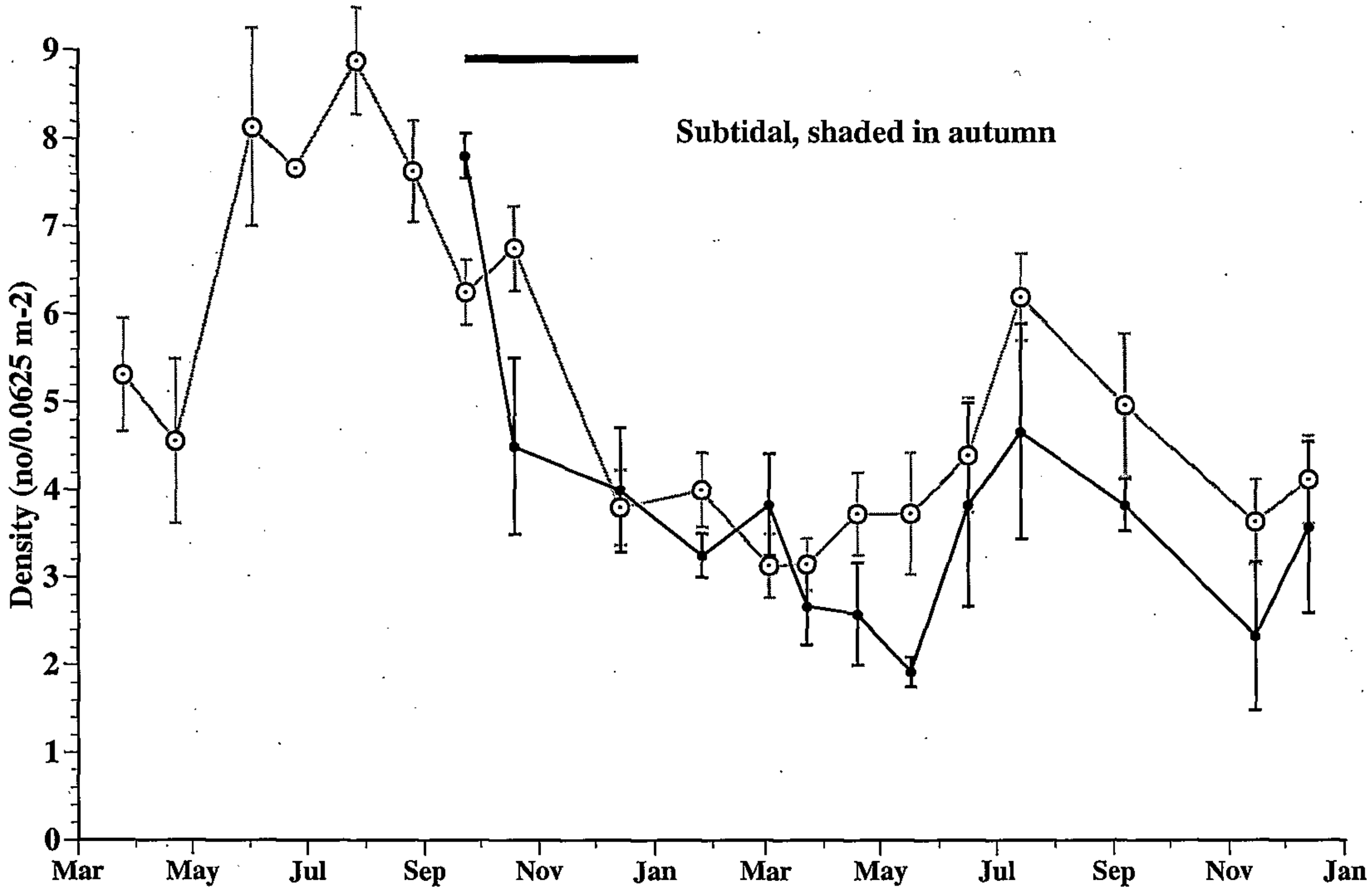


Figure 18. Density of Zostera marina at the subtidal site during 1993 and 1994 in plots (n=3) shaded for 12 weeks during autumn (solid bar) and in control plots (n=3 to 12) from all experiments; mean ± s.e.

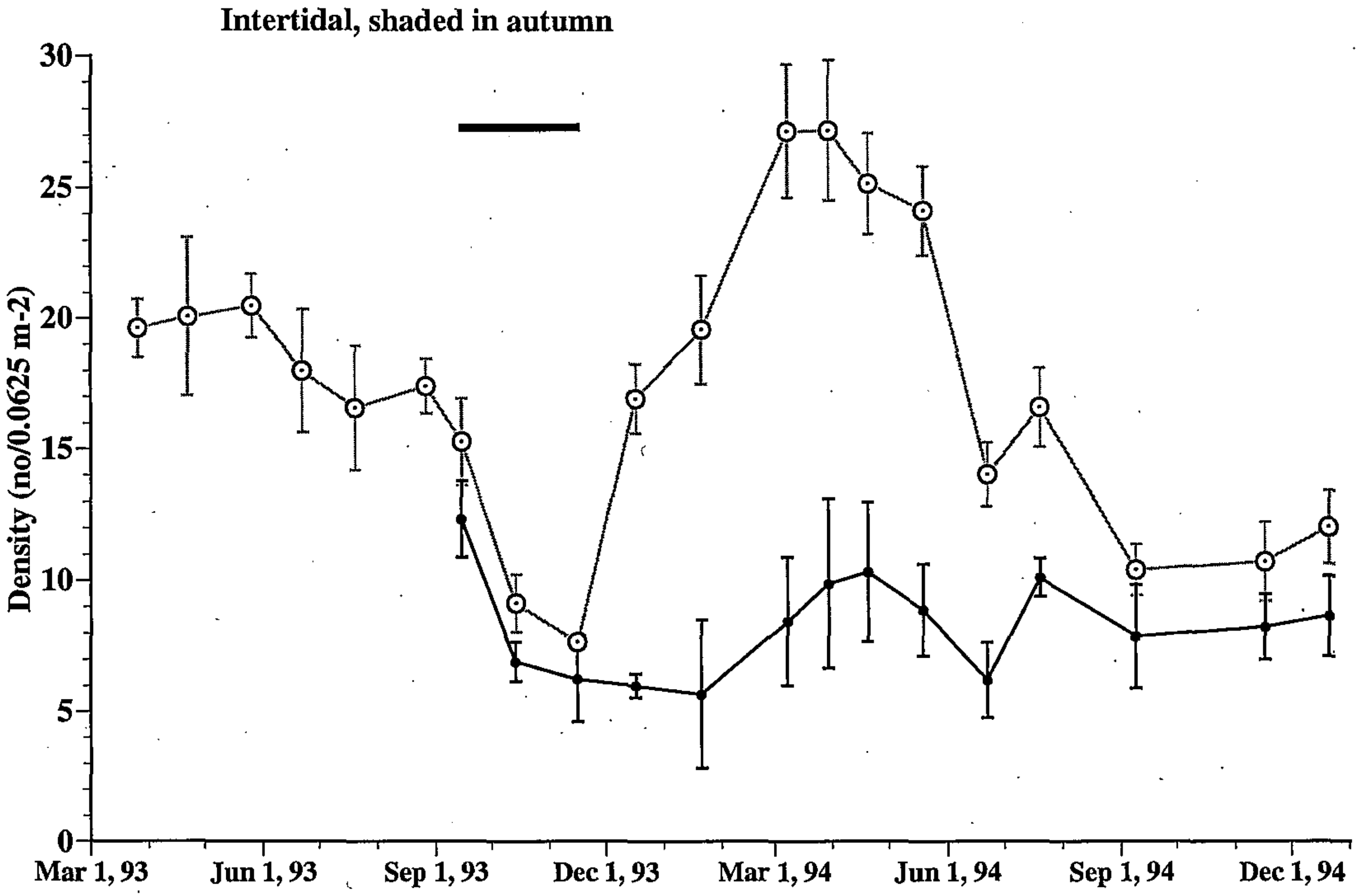


Figure 19. Density of Zostera marina at the intertidal site during 1993 and 1994 in plots (n=3) shaded for 9 weeks during autumn (solid bar) and in control plots (n=3 to 12) from all experiments; mean ± s.e.

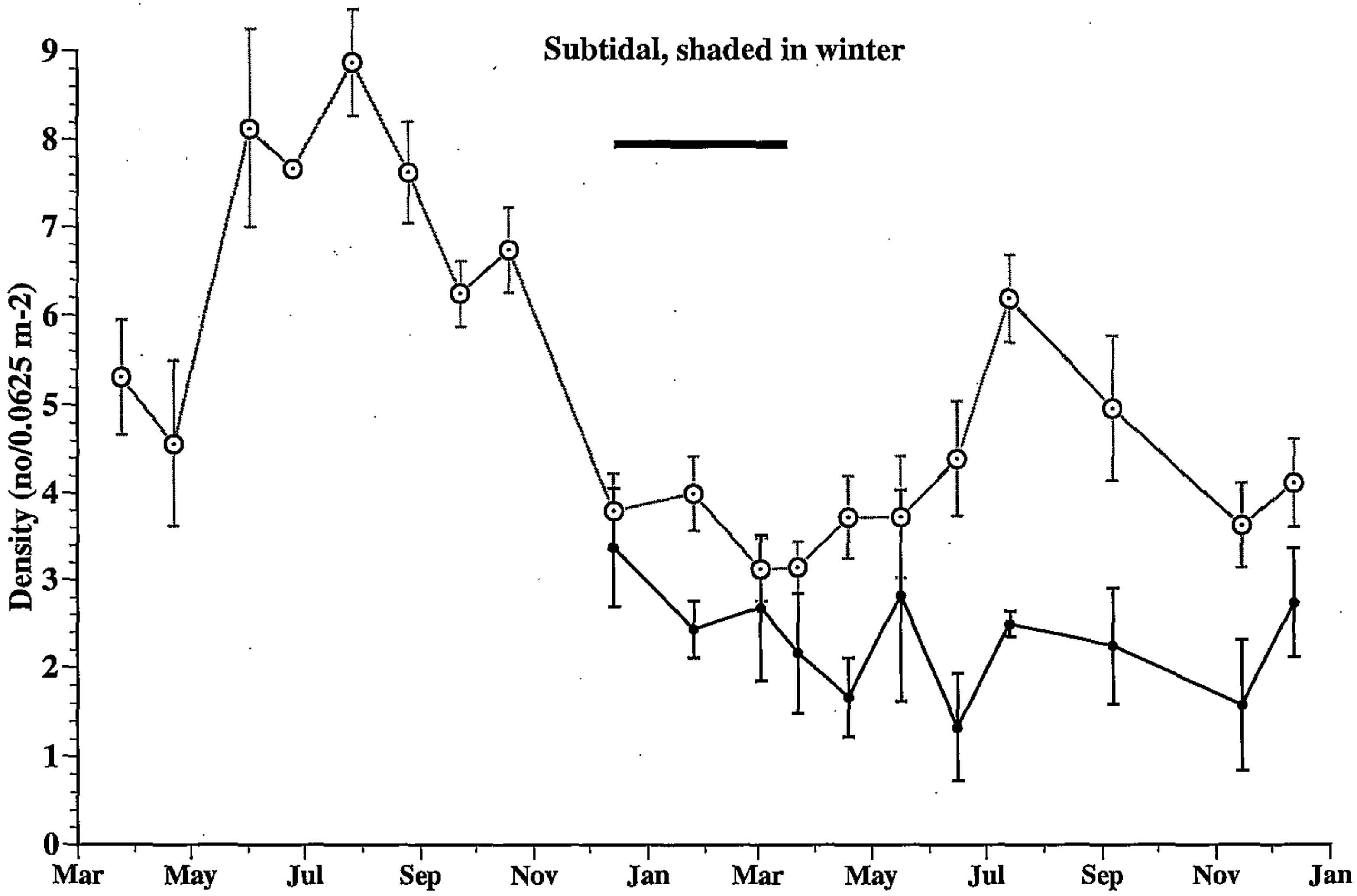


Figure 20. Density of Zostera marina at the subtidal site during 1993 and 1994 in plots (n=3) shaded for 10 weeks during winter (solid bar) and in control plots (n=3 to 12) from all experiments; mean ± s.e.

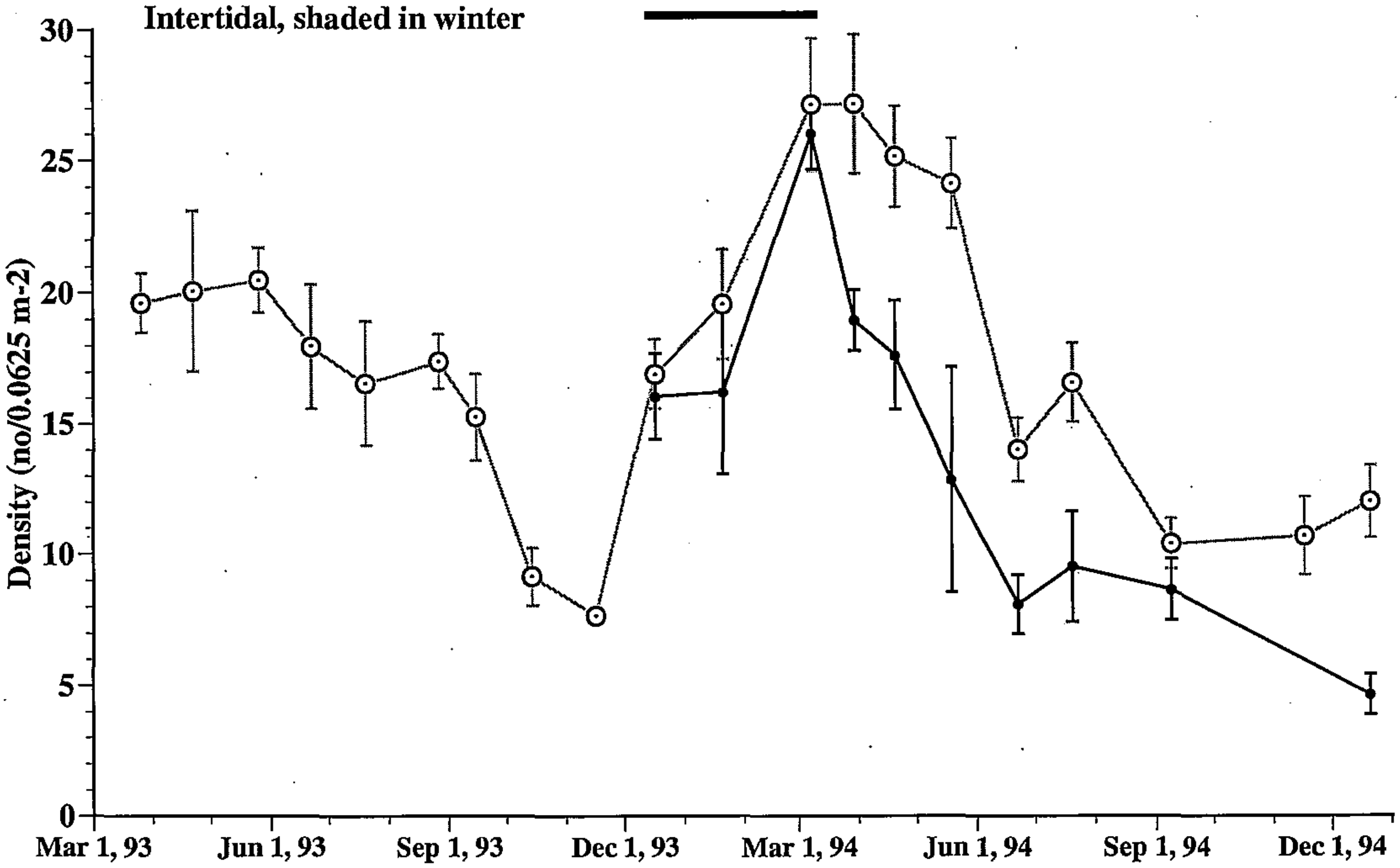


Figure 21. Density of Zostera marina at the intertidal site during 1993 and 1994 in plots (n=3) shaded for 11 weeks during winter (solid bar) and in control plots (n=3 to 12) from all experiments; mean ± s.e.