ALGAL EFFECTS ON

LITTORELLA UNIFLORA (L.) ASCHERSON

IN SCOTTISH LOCHS

A thesis submitted to the University of Glasgow for the degree of Doctor of Philosophy.

Susan J. Marrs

ProQuest LLC.
789 East Eisenhower Parkway
P.O. Box 1346
Ann Arbor, MI 48106 – 1346

DECLARATION

I hereby declare that this thesis is composed of work carried out by myself unless otherwise acknowledged and cited and that the thesis is of my own composition. The research carried out in the period October 1989 to September 1992. This dissertation in whole or in part has not been previously presented for any other degree.

Susan J. Marrs

ACKNOWLEDGEMENTS

This project was funded by the Science & Engineering Research Council in the United Kingdom and would have been impossible without the help of a large number of people. I would like to thank Professor R. Cogdell for allowing me to use the facilities of the Department of Botany, Glasgow University and for his continuous interest throughout the project. Thanks are also due to Professor M. Cowling for the use of computer facilities at Glasgow Marine Technology Centre to write the thesis, and for his encouragement and support in this task.

The project was supervised by Dr Kevin Murphy to whom I am grateful for the freedom of being permitted follow my own ideas and his active promotion of my work at various scientific meetings throughout the tenure of my studentship. Physiological work was carried out under the supervision of Dr Peter Dominy and his input is appreciated. I would also like to thank Dr Steve Maberly from the Institute of Freshwater Ecology, Windermere, for several useful discussions throughout the course of this PhD. Dr James Currall provided statistical advice.

Field work was aided by the owners of the following pairs of willing hands: Vicky Abernethy; Colin Adams; Magdi Ali; Colin Brown; Kevin Duffy; Jamie Graham; Jeremy Hills; Andy Johnson; Jim McGonagle; Rab McMath; Andrew Spink and Wee Mags. Especial thanks are given to Aileen Adams without whom the SCUBA component of the field work could not have been carried out and most certainly not have been so enjoyable. I am grateful to members of the Glasgow University Sub-Aqua Club for lending various items of equipment as my own fell into disrepair throughout the field season, namely: Al, Alan, Phil, Ruth & Sean.

I am grateful to Dr Alan Barclay (Loch of Lowes Nature Reserve); Mr Nairn (Loch End House, Lake of Menteith) and Dr F. Lees (Solway River Purification Board) for enabling me to gain access to the lochs and their continued interest throughout the project.

Dr Tippett generously allowed access to Rowardennan field station on Loch Lomond and use of the following items of field and laboratory equipment are

gratefully acknowledged: Ekman grab; LICOR light metre; pHOX dissolved oxygen meter and inverted microscope.

Thanks are also due Bob Cuthbertson for help in the design and construction of the circulatory system described in Chapter 5; Jim Muckersie for assembling the algae tanks used in Chapter 6 and Eion Robertson for his assistance with the Scanning Electron Microscope (Chapter 3). Lynda Handley from the Scottish Crop Research Institute, Invergowrie, provided advice and assistant in the measurement of $\Delta^{13}C$ (Chapter 6). Anne Neville produced Figures 5.1 and 6.3. Alan Oakman produced Figure 6.1. Alan Bell photographed Figures 2.7, 3.8 & 3.9.

My parents, Anne & David Marrs, have provided support in many ways for which I am appreciative. Thanks also to Jel for reading various drafts and providing innumerable cups of tea, and to Beatrice for helping me get away from it all when needed.

Finally, I would like to thank the gentleman who gave Aileen and myself a lift to shore when our outboard broke down one breezy day on Loch Lomond in March 1991.

Summary

In order to investigate algal effects on submerged macrophytes, four lochs experiencing a range of algal loadings were studied over a two year period. Algal loading ranged from virtually no algal growth to extensive formation of filamentous algal mats and phytoplankton blooms

During 1990 and 1991 SCUBA techniques were employed to regularly monitor the standing crop of aquatic macrophytes along the 1m isobath. In 1990 phytoplankton density was estimated by measuring phytoplankton chlorophyll *a*. In 1991 measurements of filamentous algal biomass and cover estimates of epiphytes on the leaves of *Littorella uniflora* (L.) Ascherson were also carried out.

Detrended Correspondence Analysis (DCA) of the macrophyte communities in the four lochs showed the lochs were examples of one community type. A total of 15 species were recorded, of these, the following were present in each of the four lochs: *Isoetes lacustris* L.: *Myriophyllum alterniflorum* DC: *Lobelia dortmanna* L.: *Littorella uniflora.* Conditions in the four lochs ranged from acidic Loch Dee, to the oligotrophic mid-basin of Loch Lomond, through to mesotrophic/eutrophic Lake of Menteith and eutrophic Loch of Lowes.

Morphological and some physiological attributes of *Littorella* field populations, measured over the two year period, were related to algal loading and measured abiotic parameters using stepwise linear multiple regression. Variation in morphology of *Littorella,* explained by the selected variables was generally low: this was attributed to a lack of morphological plasticity in this species. In contrast, physiological attributes, such as chlorophyll content and shoot nitrogen, had a far higher percentage variation that could be explained by the selected variables.

The abiotic variables most commonly selected, to explain variations in *Littorella* attributes, were sediment organic content and exposure rating. Sites that were more exposed to wind/wave action tended to have fewer, smaller *Littorella* plants. Sites with a high sediment organic content tended to have a greater total macrophyte and *Littorella* biomass.

Filamentous algal biomass was the most important variable that explained the variation in *Littorella* field measurements. *Littorella* plants under filamentous algal mats had a higher total leaf chlorophyll and nitrogen content and a lower chlorophyll *a:b*, indicating a possible shade response. In the presence of filamentous algal mats, *Littorella* also tended to have fewer leaves and a greater number of stolons per plant.

In the absence of filamentous algal biomass data, phytoplankton chlorophyll *a* was more commonly selected to explain variation in *Littorella* field measurements. It is suggested that in situations where filamentous algae are present, but not quantified, the effects of phytoplankton on aquatic macrophytes may be overestimated. Epiphyte percentage cover was not selected to explain any of the variation in measured field attributes of *Littorella.*

In a greenhouse experiment, *Littorella* showed a quadratic response to sediment organic content with maximal biomass accrual occurring at a sediment organic matter concentration of 75%.

After six weeks under shade conditions in the greenhouse, *Littorella* plants were significantly smaller than unshaded controls. After the removal of shading *Littorella* plants grew rapidly, until 17 weeks after the removal of shading there was no significant difference in the biomass of previously shaded plants when compared with unshaded controls. In comparison with unshaded controls, plants that had been previously subjected to shading, tended to produce fewer shorter leaves and a greater number of new plants.

Significant increases in the total chlorophyll concentration of leaves were observed 9 days after the application of shading. A decrease in chlorophyll *a:b* was only observed in experiments where shading was applied for between 3 and 6 weeks.

Photosynthetic light response curves, measured using a gas phase oxygen electrode were determined in further greenhouse experiments. These data showed shade adapted *Littorella* to have a lower maximum photosynthetic rate and higher photosynthetic efficiency at low irradiances, when compared with

unshaded controls. There were no differences between the dark respiration rates of shaded and unshaded *Littorella*. Shaded *Littorella* plants showed a higher $\Delta^{13}C$ in comparison with unshaded controls, indicating a reduction in CAM at low irradiance.

A model of population maintenance of *Littorella* in Scottish lochs is proposed. It is suggested that *Littorella* can withstand periods of algal shading by adapting physiologically with little or no loss in biomass, and the population is maintained by winter and spring growth. Loss of a population could occur under one of two sets of conditions:

1: Due to the seasonal presence of filamentous algal mats, the population becomes weakened by successive shading experiences and gradually declines.

2: The population may be able to be maintained under conditions of algal loading; however in the event of a catastrophic destruction of part or all of the population, recolonisation is not possible.

Table of Contents

List of Tables

List of Figures

Chapter 1a:

The Impact of Acidification and Eutrophication on Aquatic Macrophytes in Freshwater Lentic Systems

Chapter 1a:

The Impact of Acidification and Eutrophication on Aquatic Macrophytes in Freshwater Lentic Systems

Section 1
INTRODUCTION

There are many types of lentic system in Britain, all having one common feature in that they lack a dominant unidirectional flow (Jeffries and Mills, 1990) which makes a very specialised habitat for plant growth.

Traditionally the aquatic environment was considered to be favourable for plant growth, as the water provided physical support and protection from harsh extremes encountered by terrestrial plants resulting in an environment "peculiarly tolerable to growth and multiplication" (Lawrie, 1902). However the properties of water in comparison with air require a special set of adaptations for successful plant growth (Sculthorpe, 1967; Wetzel, 1988).

Section 2
PHYSICAL PROPERTIES OF WATER AND EFFECTS ON MACROPHYTE DISTRIBUTION

Compared with air, water is a relatively dense incompressible medium that exerts a force from all directions on any object that is immersed in it (Burns & MacDonald, 1975). Aquatic plants therefore lack the extensive support structures found in terrestrial plants, but often have gas spaces between the cells to provide buoyancy (Sculthorpe, 1967). This tissue is called aerenchyma.

Atmospheric pressure is in the region of about 1 bar, although this will vary with weather conditions and altitude. As an object goes deeper into a column of water the pressure exerted on it increases by 1 bar with every increase in depth of 10m. So, at a depth of 10m, the pressure exerted on an object will be double that exerted at the surface (Burns & MacDonald, 1975). Several authors have shown pressure

to have little effect on growth of macrophytes (Spence, 1982; Bodkin *et al.*, 1980, Peñuelas, 1988). Wetzel (1988) suggests pressure may act in concert with light and temperature to limit macrophyte colonisation to 10m, although there are examples of macrophyte colonisation exceeding this depth (e.g. Spence *et al.*, 1979).

The action of water movement caused by wind/wave action controls the distribution of sediments (Allen, 1985) and so affects the distribution of macrophytes. Waves are formed by the turbulence caused by fast moving air flowing over the surface of water.

The size of particles in an area of lake substrate depends on the critical erosion and settling velocities of the particle. Smaller particle sizes are deposited in areas of low velocity. Once suspended in the water column, particles are sorted by a combination of wave action, turbulent mixing and longshore currents. More dense particles are deposited first, resulting in sediment becoming finer with increasing distance from the lake edge and increasing depth.

In oligotrophic lakes, sites that are exposed to a greater wind/wave action may have a lower productivity than more sheltered sites. Emergent macrophytes will be distributed closer to the shore in more exposed sites when compared to sheltered sites (Keddy, 1983). Weisner (1987) reported reduced macrophyte growth in the more sheltered regions of a eutrophic lake. In both cases reduced macrophyte growth can be related to sediment type and wave exposure.

In oligotrophic lakes, low nutrient availability and mechanical damage may serve to limit macrophyte growth in more exposed areas (Keddy, 1983). Where the substrate is rocky, macrophyte colonisation may not occur as roots are unable penetrate the substratum (Spence, 1982). In the sheltered area of a eutrophic lake, anoxic sediments may develop resulting in reduced macrophyte growth (Smits *et al.*, 1990). In very sheltered conditions the sediment may become unstable, and soft, resulting in macrophytes being unable to establish effective anchorage (Spence, 1982).

The influence of sediment type on the growth and colonisation of aquatic macrophytes is considered in Chapters 2 and 3, where the physical aspects of the sites in relation to their flora are discussed. In Chapter 5 the effect of sediment type is further considered with a greenhouse experiment investigating the effect of sediment organic matter content on the growth of *Littorella uniflora* (L.) Ascherson.

Section 3

PHOTOSYNTHESIS IN THE AQUATIC ENVIRONMENT

3.1 Light in Lakes

Light is the major factor influencing depth distribution of aquatic macrophytes (Spence, 1982; Spencer & Bowes, 1990; Duarte, 1991). Usually the limit of macrophyte growth is within a few metres of the surface with dissolved organic matter and suspended particles rapidly absorbing and scattering any incident light (Kirk, 1983). However, in clear water lakes macrophyte growth can extend far deeper, for example Loch Borraile near Durness in Sutherland (Scotland) has dense swards of *Nitella* at depths of 12 to 15 metres (Spence *et al.*, 1979). This loch is considered to be one of the lakes most deeply colonised by macrophytes in Britain.

In terms of light quantity, a lake can be divided into two zones. In the upper reaches of the water column light intensities are such that photosynthesis can occur and plant life can be sustained: this region is known as the littoral zone. With increasing depth the light intensity will decrease until the point is reached where plant photosynthesis just compensates for plant respiration, known as the light compensation point. This point is the divide between the euphotic zone and the profundal zone, where light intensities are so low that respiratory losses of carbon exceed photosynthetic gains and plant life can no longer be sustained.

Three major influences will affect the passage of a photon of light through the water column: diffraction; reflection and refraction. Diffraction is caused by particles in the water column deflecting light from its course. Reflection occurs when a particle of light comes in contact with a surface resulting in a change of

direction; the light does not cross this surface. Refraction is where light enters a particle and emerges after one or more internal reflections.

A particle will scatter light if it is greater in diameter than the wavelength of incident light, so scattering of photosynthetically active radiation (400-700nm) will be selective when particles are less than 700nm, in diameter. In pure water, scattering is brought about by water molecules and is increased by the wavelength of light to the power -4 (λ-4). Blue light is therefore scattered more than red light. In clear water, light intensity decreases almost exponentially with depth. Non-exponential decay of irradiance is brought about by changes in the spectrum and vertical differences in both attenuation and scattering (Kirk, 1983).

Turbidity is caused by suspended inorganic particles such as clays and silts (tripton) scattering light and acting as neutral density filters with little effect on light quality (Kirk, 1983). In shallow lakes, where most of the lake bed is in the wave mixed zone, effects due to turbidity are at their greatest (Jackson & Starrett, 1959; Van Dijk & Achterberg, 1993).
Most natural waters are coloured due to selective absorption of light by dissolved organic matter and organic colloids, termed gelbstoff, yellow substances or gilvin. These substances absorb greatly in the ultraviolet and blue region of the spectrum and are the reason that many freshwater bodies appear brown or yellow in colour (Kirk, 1976, 1983). Light is also selectively absorbed by the photosynthetic pigments in phytoplankton and their cells will scatter light (Kirk, 1975a,b, 1976, 1983).

The resultant effect of the above is that light attenuation in water is diffuse. The vertical attenuation coefficient is a description of the scattering and absorption of light by all of the above components - its calculation is described in Chapter 2.

Schanz & Felix (1991) measured the respective contributions of gilvin, phytoplankton, water and tripton to the attenuation of light in Lake Zurich during spring 1986. They found phytoplankton attenuation to be the dominant factor in reduction of light transmittance through the water column. Van Dijk & Achterberg (1993) found tripton attenuation to be the dominant factor in light reduction in

Lake Veluwe, The Netherlands. Both studies showed water and gilvin to be of minor importance.

3.2 Carbon dioxide

Water in equilibrium with air will contain the same concentration of carbon dioxide as the atmosphere. However, slow carbon dioxide diffusion rates ($x10^4$ slower than in air) and the presence of a boundary layer around leaf surfaces often result in aquatic photosynthesis being limiting by the availability of carbon dioxide. Madsen *et al.* (1993) found photosynthetic rates to increase at elevated carbon dioxide levels in several species of aquatic macrophyte.

Carbon dioxide dissolves in water to form a weak acid - carbonic acid. A property of weak acids is that their dissociation into ions is very malleable. Dissociation of carbonic acid in water follows the following pH-dependent equilibrium equation (see Golterman, 1975):

Equation 1. **Equilibrium equation for the dissociation of carbonic acid in water.**

$$CO_2 + H_20 \leftrightarrow H_2CO_3 \leftrightarrow H^+ + HCO_3^- \leftrightarrow 2H^+ + CO_3^{2-}$$
$$\text{pK } 6.2 \qquad \text{pK } 10.3$$

The chemical composition of natural waters can shift the equilibrium of the above dissociations and different aquatic macrophyte species may adapt to utilise different forms as their source of carbon: this has important implications for habitat selection (Raven *et al.*, 1985; Boston *et al.*, 1989; Spencer & Bowes, 1990).

All aquatic macrophytes appear to have a greater affinity for carbon dioxide than bicarbonate (Boston *et al.*, 1989). In nutrient-rich alkaline sites the relative abundance of bicarbonate is much greater than that of carbon dioxide (see Equation 1) and many macrophytes inhabiting these areas have mechanisms to allow the utilisation of bicarbonate and so increase the available carbon pool (Maberly & Spence, 1983; Sand-Jensen, 1983, 1987; Jones *et al.*, 1993).

In nutrient-poor systems, there tend to be no bicarbonate users although carbon may still limit photosynthesis due to the low levels of total inorganic carbon. These systems tend to have low pH and the proportion of carbon available as bicarbonate is low (Golterman, 1975). Nutrient availability will also limit plant growth. Spence (1964) suggests that the paucity of species that overcome carbon limitation by the formation of floating leaves in nutrient-poor lakes is due to lack of available nutrients. In these systems carbon conservation and recycling is more important than competition (Boston *et al.*, 1989). Adaptations such as a thick cuticle to prevent carbon dioxide loss, recycling endogenous carbon dioxide, CAM-like metabolism which extends the effective daily period of carbon fixation, and carbon dioxide uptake from the sediment are common.

CAM-like metabolism is described more fully in Chapter 1b, where the relevance of this photosynthetic pathway is discussed in relation to the ecology of *L. uniflora.*

3.3 Oxygen

A second gas that is important to photosynthesis in aquatic systems is oxygen. The degree of solubility is dependent on temperature, with a greater amount of oxygen being dissolved in cooler water. Oxygen concentration will also vary, depending on the amount of turbulence in the water column and the relative balance between photosynthesis and respiration.

Dissolved oxygen concentration shows a marked diel fluctuation in lakes colonised by macrophytes and algae. During the day, the water column may become super-saturated (i.e. greater than 100% saturation) due to photosynthesis. However, respiration at night may cause oxygen levels to fall very low and in extreme conditions may result in large scale suffocation of fish (e.g. Townsend *et al.*, 1992).

Under conditions of high oxygen concentration, photosynthesis may become inhibited. Ribulose-1,5-bisphosphate carboxylase/oxygenase (RUBISCO) is the primary carboxylation enzyme of C3 photosynthesis. Under conditions of high carbon dioxide concentration RUBISCO catalyses the reaction between carbon

dioxide and ribulose-bisphosphate to produce 2 molecules of phosphoglycerate (PGA). PGA is then incorporated into subsequent photosynthetic reactions to produce carbohydrate and oxygen. RUBISCO will also catalyse the oxygenation of RUBP resulting in 1 molecule of PGA and 1 molecule of phosphoglycolate which may be further metabolised to produce carbon dioxide. This process is termed photorespiration and may greatly reduce the efficiency of photosynthesis (Björkman, 1981).

In terrestrial systems some species have evolved to overcome the problems of photorespiration by utilising C4 photosynthesis (see Ehleringer *et al.*, 1991). In this system carbon, is fixed as four carbon acid in the mesophyll cells in a reaction catalysed by phosphoenol pyruvate (PEP) carboxylase. The fixed carbon is then transported to the bundle sheath where decarboxylation occurs and carbon dioxide is released. Carbon dioxide fixation then proceeds as in C3 photosynthesis.

Due to the large daily fluctuation in oxygen, and a concomitant reduction in carbon dioxide, conditions that may result in photorespiration are not uncommon in aquatic systems (Simpson *et al.*, 1980; Søndergaard, 1988). Photorespiration may be reduced by the utilisation of carbon-concentrating mechanisms (e.g. Maberly & Spence, 1983; Sand-Jensen, 1983, 1987; Jones *et al.*, 1993). A system similar to C4 photosynthesis of terrestrial plants has not been discovered in aquatic systems (Salvucci & Bowes, 1983; Søndergaard, 1988).

Section 4

ANTHROPOGENIC EFFECTS

4.1 Eutrophication

Eutrophication of a water body is the process of nutrient enrichment and may occur either naturally, or due to anthropogenic influences, for example water run off from fertilised agricultural land, or from sewage effluents. Such an increase in nutrients may have a marked effect on the flora of the affected water body such as a decrease in species diversity and changes in the dominant flora (Arts *et al.*, 1990a; Lachavanne, 1985).

The simplest hypothesis for the loss of macrophyte species with eutrophication is that offered by Wetzel and Hough (1973), where with increased nutrients, phytoplankton are able to grow more rapidly, and macrophytes are reduced due to shading effects of phytoplankton. Mulligan and Baranowski (1969) observed that with a continued loading of nutrients in a set of study ponds, dominance progressed through a succession of macrophytes to filamentous algae to phytoplankton. An increase in filamentous algal biomass with increasing nutrient input has been observed by other workers in lentic systems (Howard-Williams, 1981) and in riverine natural and artificial systems (Spink, 1992).

The hypothesis of Phillips *et al.* (1978) suggests that in the non-enriched situation macrophytes compete with phytoplankton for nutrients and secrete allelopathic substances that reduce phytoplankton growth. As available nutrients in the water column increase, epiphytic growth increases with a resultant decrease in macrophyte growth. Decreased macrophyte growth leads to a reduction in the production of allelopathic substances, and a consequent increase in phytoplankton growth. The increase in phytoplankton will further reduce macrophyte growth due to competition for light so resulting in a positive feedback mechanism with increasing phytoplankton growth and decreasing macrophyte growth.

Hough *et al.* (1989) modified the hypothesis of Philips *et al.* (1978) to include effects of non-rooted macrophytes. These authors carried out a study on a chain of

water bodies with similar watershed to determine the community dynamics of the flora over a range of nutrient status.

Glass slides colonised by epiphytes, to a density equivalent to that found on macrophyte leaf surfaces in the field, absorbed 84% of incident light, demonstrating that epiphytic growth can result in a great reduction in PAR available to macrophytes (Philips *et al.*, 1978). Sand-Jensen (1977) showed that diatom crusts on eel grass greatly reduced levels of light and acted as a barrier to bicarbonate diffusion.

Epiphytes and phytoplankton may respond equally to an increase in nutrients, but as fewer individual epiphytic cells are lost than phytoplankton there may be an apparently greater increase in biomass of epiphytes. Losses in phytoplankton are caused by sedimentation and grazing, whereas the major cause of loss of epiphytes is by of old plant leaves. In some stratified lakes where the epilimnion is deeper than the photic zone, phytoplankton may be circulated out of the photic zone, whereas epiphytes are maintained in a more constant light regime which may result in a more favourable productivity (Sand-Jensen & Søndergaard, 1981).

Simpson & Eaton (1986) compared the photosynthesis and respiration of *Elodea candensis* and two species of filamentous algae. *E. canadensis* experienced greater inhibition of photosynthesis under conditions of high oxygen concentration than the two algal species. Photosynthetic and respiratory measurements were also carried out over a range of pH and carbon dioxide concentration; again *E. canadensis* showed a greater reduction in photosynthesis at high pH and low carbon dioxide concentration. These workers suggest that the ability of the two species of filamentous algae to photosynthesis more efficiently under conditions of high oxygen, low carbon dioxide and high pH confer a competitive advantage over *E.canadensis*. This, in combination with the shading effects of filamentous algae, could result in the loss of aquatic macrophytes from enriched aquatic systems.

There is evidence that the presence of macrophytes has an adverse effect on the growth of phytoplankton. Reduced densities of phytoplankton have been observed in the water column above macrophyte beds (Hasler & Jones, 1949; Barko *et al.*,

1988). There are three main possible causes of this. Losses could be incurred in the phytoplankton population by: increased settling due to lack of turbulence: secretion of allelopathic substances: competition for nutrients with the macrophytes: macrophytes providing a habitat for grazing invertebrates.

Evidence for secretion of allelopathic substances is scarce (Sand-Jensen & Søndergaard, 1981). Most studies have tried to find possible allelopathic substances in plant extracts that may not be exuded. Allelopathy has been recorded in freshwater and brackish Characeae with a resultant suppression of both epiphytes and phytoplankton (Wium-Andersen *et al.*, 1982; Hootsmans, 1991). At present there is no consensus of opinion on the regulation of phytoplankton community dynamics in macrophyte beds (Barko *et al.*, 1988). Algal growth measurements in filtered and nutrient enriched water, sampled from within macrophyte beds, produced some evidence of allelopathic effects (Hootsmans, 1991). These results were not conclusive, as reduced growth of *Scenedesmus* was observed in some samples, but not in others.

Grazing effects by zooplankton has a significant effect on the algal population in aquatic systems. Using minimal models, Scheffer (1991) demonstrated that gradual changes in the density of planktivorous fish could lead to quite rapid changes in both zooplankton and phytoplankton. In conditions where nutrient conditions were increased in the absence of planktivorous fish, the model predicted that numbers of zooplankton would increase, whereas those of phytoplankton would remain low due to grazing.

Schoenberg (1990) demonstrated that a reduction in zooplankton resulted in an increase in phytoplankton in short-term enclosure experiments. Stansfield *et al.* (1989) presented evidence that the loss of macrophytes from the Norfolk Broads coincided with the presence of high levels of pesticides such as dieldrin and polychlorinated biphenyls resulting in a the death of phytoplanktivorous zooplankton such as *Daphnia.* On the basis of these observations, experimental work was carried out concerning the use of phytoplanktivorous species such as *Daphnia* in the amelioration of aquatic systems suffering from extensive algal growth (Irvine *et al.*, 1990; Moss *et al.*, 1991). Provision of refuges for

zooplankton in order to reduce predation by planktivorous fish proved unsuccessful (Irvine *et al.*, 1990). Enclosure experiments demonstrated that in order for *Daphnia* species to survive it was necessary to reduce predation and that a suitable food source was available (Moss *et al.*, 1991). These workers concluded that biomanipulation of eutrophic water bodies could be achieved by encouraging the growth of *Daphnia* communities

Other important factors that may contribute to macrophyte loss are: putrid silt formation due to organic enrichment: mechanical effects: competition for light and/or nutrients: epiphytic algae on small macrophyte shoots in the deepest zone (Lachavanne, 1985).

4.2 Acidification

In many regions of the world the acidity of lakes and rivers has increased since the beginning of the industrial revolution. In Scotland, studies on diatom species assemblages in core samples collected from lakes showed a change occured circa 1850 in species from those commonly found in habitats of a pH around 7 (circumneutral) to those found in more acid conditions (acidophilous). A further shift in diatom species was observed around the turn of the century with diatom species that preferably inhabit loch water around pH 5.5 or less (acidobionts) being observed. In the absence of any adequate alternative hypothesis, the primary cause of this reduction of pH was concluded to be acid deposition (Battarbee, 1984; Battarbee *et al.*, 1985). Upland afforestation, especially where species such as Sitka spruce are present has been demonstrated to exacerbate this situation (Harriman & Morrison, 1982).

Freshwaters susceptible to acidification are those which run over base-poor rocks such as granite or gneiss. They are usually already low in nutrients, with very clear water due to lack of dissolved organic matter and are not to be confused with naturally acidic waters in peat rich areas that are discoloured due to the high humic acid content of the water (Cresser *et al.*, 1988).

Acid waters typically have low levels of HCO_3^- and consequently have a greatly reduced buffering capacity to any incoming acid components. There is some

discussion as to whether increased acidification leads to oligotrophication; nutrient input may not decrease, but could increase due to increased atmospheric nitrate loadings. Phosphate has been shown to co-precipitate with aluminium at low pH, thus rendering it unavailable for assimilation by primary producers (Conway & Hendry, 1982; Morrison & Batterbee, 1988). Neuvonen & Suomela (1990) observed reduced decomposition rates of birch and pine needles under simulated conditions of acid deposition in forests in northern Finland. Evidence for reduced decomposition rates in acid conditions is backed up by observations of large amounts of undecomposed allochthonous matter in some acid lakes (Leivistad *et al.*, 1976)

Enclosure experiments carried out in Quebec, Canada (Delisle *et al.*, 1984) showed reduced pH to have a profound effect on phytoplankton. Enclosures treated with sulphuric acid showed a reduction in the number of species, and a switch from cyanobacteria to Chlorophyceae in comparison to untreated controls. Low species diversity in acidified lakes has been well documented (Chrisman *et al.*, 1980; Conway & Hendrey, 1982; Lyden & Grahn, 1985, Ilmavitra, 1988).

Other enclosure experiments have suggested that reduction in phytoplankton growth in acidified lakes is not due to acidification alone (Yan & Stokes, 1987). There is evidence that decreased phytoplankton growth may be brought about when levels of aluminium in the waters are increased (Nalewajko & Paul, 1985), possibly due to co-precipitation with phosphate rather than any direct toxic effects of aluminium (Morrison & Battarbee, 1988).

Not all algal groups show an acidity induced reduction in number. At low pH extensive filamentous algal mats of species such as *Mougeotia* spp are common (Hendrey & Vertucci, 1980; Lazarek, 1985; Raven, 1988).

Low pH has been shown to result in reduced seed production in *Najas flexilis* (Titus and Hoover, 1993). In the same study *Vallisneria americana* failed to flower and produced fewer tubers at low pH. These authors introduced the 'closing spiral' hypothesis whereby plants grown at low pH show reduced growth and

reproductive success resulting a spiralling decrease in population size through successive growing seasons.

In Scandinavia, The Netherlands, the British Isles and the United States similar patterns of changes have been recorded for lake macrophyte flora in response to acidification, with an increase in *Sphagnum* spp. and *Juncus bulbosus* and a decrease in isoetids (Roelofs, 1983; Gran, 1985; Arts *et al.*, 1990b; Farmer, 1990; Morris, 1991).

Roelofs *et al.* (1984) attributed changes in the flora of acidified lakes to changes in carbon budget. When photosynthetic carbon response curves of *L. uniflora* and *J. bulbosus* were compared, both species showed an increase in photosynthetic rate with increasing carbon levels, up to levels of 2mM and 0.5mM respectively. However, *J. bulbosus* exhibited greater photosynthetic rates at all carbon levels. In more acid waters free carbon dioxide increases in concentration, possibly due to mobilisation in the sediment. Roelofs *et al.*, (1984) found that *J. bulbosus* took up carbon dioxide from the shoots only, whereas *L. uniflora* takes up most carbon via the roots (Søndergaard & Sand-Jensen, 1979), so acidification increased carbon source for *Juncus*, but not for *Littorella.*

Experimental liming of acidified lakes has brought about a reverse in the floristic changes due to acidification at several sites in Sweden (Eriksson *et al.*, 1983; Farmer, 1990) and in Britain (Battarbee *et al.*, 1991; Marrs *et al.*, 1993).

Section 5:
MACROPHYTE SPECIES STUDIED

In order to investigate algal effects on aquatic macrophytes it was decided to concentrate on one species. The decline of isoetids with both acidification and eutrophication has been well documented (e.g. Arts *et al.*, 1990b; Farmer, 1990). Of all the isoetids, *Littorella uniflora* (L.) Ascherson has the widest distribution (Farmer & Spence, 1986) and suitable populations were readily located in a range of habitats, ranging from acidified to eutrophic. Chapter 1b gives an account of the ecology and physiology of *L. uniflora* (hereafter *Littorella*).

Chapter 1b:

The Ecology & Physiology of *Littorella uniflora* (L.) Ascherson

Chapter 1b:

The Ecology and Physiology of *Littorella uniflora* (L.) Ascherson

Littorella uniflora is a plant native to Britain and can typically be found on exposed lake shores and in shallow fresh water. Although once common in suitable habitats throughout the British Isles up to an altitude of 826m (Wilson, 1949), *Littorella* is currently on the decline in lowland areas (Farmer & Spence, 1986; Stace, 1991); its decline has also been noted in European waters (e.g. Arts *et al.*, 1989 & 1990b; Roelofs, 1983; Roelofs & Schuurkes, 1983; Szmeja, 1989; Voëge, 1989).

Littorella's compact, rosette growth-form is typical of the isoetids to which it belongs. The isoetids are a small group of morphologically similar, but taxonomically unrelated, aquatic macrophytes typical of soft waters. Other members of this group common in Britain include *Subularia aquatica* L., *Lobelia dortmanna* L., *Isoetes lacustris* L. and *Isoetes echinospora* Durieu. As well as a compact, rosette growth form, isoetids are characterised by having stiff green leaves; a high root to shoot ratio in terms of surface area, weight and volume; and a high percentage of the internal volume being taken up by gas-filled lacunae (Den Hartog & Segal, 1964; Boston, 1986; Raven *et al.*, 1988).

Littorella is the only member of the Plantaginaceae to produce unisexual flowers. Flowers are produced during the months of June to August (Clapham *et al.*, 1987) only when the plant is exposed to the atmosphere, with female flowers developing at the base of the male scape (Stace, 1991). On fertilisation a single achene is produced which has a length of 2mm and a weight of 1mg (Swarbrick & Raymond, 1970). Reproduction and colonisation are, however, primarily by growth of stolons (Stace, 1991). Arts and Van der Heijden (1990) have demonstrated that optimum seed germination (76%) occurs after a period of drying (2-4 weeks), followed by waterlogging. This may be a useful adaptation in allowing *Littorella* to colonise previously exposed inundated land more rapidly than the growth of stolons alone would allow.

Littorella typically inhabits lakes with sandy or gravel sediment of a low inorganic content (Roelofs, 1983) and pH range of 5 - 8 (Misra, 1938). Most individuals grow on sediments that are sub-optimal for growth, possibly due to their compact growth form which renders them unable to extend in height above any sedimentation that may occur in richer water bodies (Wilson & Keddy, 1985). The plants can withstand reasonable amounts of disturbance from wave action due to their large root systems which act as effective anchorages (Keddy, 1983; Spence, 1982). Large root systems also provide a large surface area for nutrient and sediment carbon dioxide uptake (Søndergaard & Sand-Jensen, 1979).

In northern Polish oligotrophic lakes *Littorella* was found to be a major competitor with *Lobelia dortmanna,* with *Littorella* being more able to recolonise disturbed areas than any other species present. Complete recolonisation of cleared sites took five years and the primary coloniser was *Littorella* (Szmeja, 1987). However, when conditions are more favourable for the growth of other species, *Littorella* may be rapidly out-competed by species such as *Potamogeton polygonifolious* Pourett (Guppie, 1917), *Myriophyllum alterniflorum* DC or *Ranunculus peltatus* Schrank (Roelofs, 1983). When sufficient nutrients are available, growth of *Littorella* is rapid; however, plants can survive for longer than six months with no change in appearance when nutrient or light levels are low (Holstrup & Wiegleb, 1991b).

The depth distribution of *Littorella* ranges from exposed on water-logged soil, down to a depth of 4m (Stace, 1991), although frequently at a depth of around 2m *Littorella* is replaced by *Isoetes lacustris* (Sand-Jensen & Søndergaard, 1979). *I lacustris* fares better at lower light levels than *Littorella* by having a lower dark respiration rate, but is outcompeted by *Littorella* at higher light levels (Sand-Jensen, 1978). Farmer & Spence (1986) found *Littorella* plants from deeper water to have larger leaf surface areas, a lower light compensation point, a higher chlorophyll content and a lower chlorophyll *a:b* ratio than shallow water plants. Data from Søndergaard & Bonde (1988) agree with this fairly typical example of shade adaptation.

Submerged leaves of *Littorella* do not survive exposure to the air for long periods of time, and within five days new aerial leaves may be formed. If the ground is dry these leaves lie flat on the sediment, but on the onset of waterlogging soon stand almost upright. Should the plant be resubmerged the aerial leaves can survive for several months, and after several weeks it is impossible to tell if a submerged leaf emerged when the plant was exposed or under water (Holstrup & Wiegleb, 1991b). Architecture of both leaf types are similar, however aerial leaves have numerous stomata that are not present on submerged leaves (Aulio, 1985; Holstrup & Wiegleb, 1991b). In submerged leaves the central vascular bundle is open to the surrounding water - this is closed in aerial leaves (Holstrup & Weigleb, 1991a). Pedersen & Sand-Jensen (1993) demonstrated this morphology to be related to acropetal water transport in the absence of transpiration. This phenomenon has been quantified in *L. dortmanna* and *Sparganium emersum* Rehman (Pedersen, 1993).

In common with several, but not all, isoetids, *Littorella* shows several adaptations to optimise carbon acquisition and utilisation, which may be of extreme importance in aquatic systems where competition for carbon is fierce (Boston, 1986; Bowes, 1987; Raven *et al.*, 1988). In a comparison with other submerged macrophytes, Madsen *et al.* (1993) found isoetids to have a low photosynthetic rate relative to the activity of carbon-fixing enzymes.

In oligotrophic waters, where photosynthesis may be carbon limited, carbon dioxide concentrations in sediment interstitial water may be as much as 100 to 400 times greater than that of the overlying waters (Wium-Andersen & Andersen, 1972). Field experiments with $^{14}CO_2$ have demonstrated that primary uptake of inorganic carbon by *Littorella* is via the roots, with up to 95% of daily carbon uptake from the sediment (Søndergaard & Sand-Jensen, 1979). Although more carbon is available to plants due to utilisation of sediment carbon dioxide, levels are still often below those necessary to maximise photosynthesis, and growth is still likely to be carbon-limited (Boston, 1986).

It has been demonstrated that the roots of *Littorella* have a high permeability to oxygen, and this has been suggested as one of the reasons *Littorella* is not found on

highly anoxic sediments (Smits *et al.*, 1991). Root oxygen evolution is high in comparison to non-isoetid species: *circa* 30% and 4% for *Littorella* and non-isoetids respectively (see Raven *et al.*, 1988).

Littorella exhibits Crassulacean Acid Metabolism (CAM) (Boston & Adams, 1983) whereby respired carbon dioxide is refixed due to the activity of the enzyme phosphenolpyruvate (PEP)-carboxylase and is stored in the vacuole as malic acid during the night. During periods of illumination (daytime), carbon dioxide from malic acid is incorporated into the normal C3 photosynthetic pathway (Stryer, 1981). This photosynthetic adaptation is normally associated with terrestrial plants in arid regions that are required to minimise water loss (Ting and Rayder, 1982), but is also utilised by some aquatic species under conditions where inorganic carbon may be limiting (see Bowes, 1987; Raven *et al.*, 1988) as the diel period for carbon assimilation is effectively lengthened (Spencer & Bowes, 1990).

Due to the extra enzymatic reactions required for CAM, carbon fixation is expensive in terms of ATP and NADPH compared with the C3 meachanism (Nobel, 1991). Under conditions of high carbon dioxide concentration during daylight, diurnal fluctuations of malate do not occur, i.e. CAM does not occur. Accumulation of malate in the vacuole at night is greatest under conditions where daytime levels of carbon dioxide are low and night levels are high (Madsen, 1987b; Holstrup & Wiegleb, 1991b). Boston & Adams (1986) found CAM to contribute 45 to 55% of the annual carbon gain for populations of *Littorella uniflora* var. *americana* in softwater lakes in Wisconsin. Terrestrial life forms of *Littorella* do not express CAM (Aulio, 1985) unless conditions of 100% relative humidity prevail, a rare condition in nature (Aulio, 1986).

A final adaptation of *Littorella* to low carbon levels is the ability to recycle carbon dioxide that has been respired into the lacunae. Fifty-to-70% of carbon dioxide expired during the day can be refixed (Søndergaard, 1979) by the chloroplasts which are situated in the tissues lining the lacunae of *Littorella* (Raven, 1970; Sculthorpe, 1967). Recycling of significant amounts of carbon dioxide is only possible in species with extensive lacunae (Søndergaard, 1979).

Boston and Adams (1986) have produced the following model of the carbon budget in Littorella.

Fig 2.1 Model of carbon budget for *Littorella uniflora.*

NET		CAM	C3
ANNUAL	=	NOCTURNAL CARBON +	DAYTIME CARBON —*LOSS
PRODUCTION		FIXATION	UPTAKE

*Losses were estimated to be respiration + photorespiration + secretion + grazing - daytime refixing of endogenous carbon dioxide.

Chapter 1c:

Aims & Approaches

Chapter 1c:

Aims & Approaches

In lakes where either acidification or eutrophication is taking place there is a change in the balance between macrophytes and algae. Both scenarios may result in the loss of macrophytes, or a reduction in macrophyte species diversity. Few studies have considered algal/macrophyte relationships over a range of conditions from eutrophic to acidified.

In both scenarios, competition for carbon between macrophytes and algae will be fierce (Maberly & Spence, 1983), and oxygen production by filamentous algal mats in eutrophic systems has been shown to have detrimental effects on elodeids (Simpson & Eaton, 1986). Again in both systems mechanisms for increasing the available carbon have been identified, however quite different mechanisms are employed in the two different situations (see Søndergaard, 1988).

There were three main aims of this thesis:

1. To determine the effect of algal growth on *Littorella* in both acidified and eutrophic lakes.

2. There are three main algal components in freshwater lakes: epiphytes, filamentous algae and phytoplankton (see Figure 3.1 p46). The second aim of this thesis was to investigate which of these algal components had the greatest effect on *Littorella.*

3. *L. uniflora* takes up the bulk of its carbon via the roots (Søndergaard & Sand-Jensen, 1979) and has been demonstrated to have a high permeability to oxygen leakage through the roots (Smits *et al.*, 1991). Root oxygen evolution is high in comparison to non-isoetids: *circa* 30% and 4% for isoetids and non-isoetids respectively (see Raven *et al.*, 1988). Due to the high levels of gaseous exchange through the roots of *Littorella*, effects due to carbon competition and increased oxygen concentration are caused by algal photosynthesis on the plant surface and in

the surrounding water are likely to be minimal, in comparison with those published for elodeids (e.g. Simpson & Eaton, 1986). As a consequence of this, it was decided to concentrate the study on the possible effects of shading by algae, and the third aim of this work was to determine both the morphological and physiological adaptations of *Littorella* to shade which may be brought about by the presence of algae.

Two main approaches were adopted:

1. **Hypotheses generation** (Chapters 2 to 4)
Four lochs, that experienced a range of algal loading, were surveyed over a two year period. Loading estimates of phytoplankton, epiphytes and filamentous algal mats, along with abiotic parameters were regressed against field attributes of *L. uniflora*. Hypotheses regarding changes in morphology and physiology with algal loading were derived using the results of these regressions.

2. **Hypothesis testing** (Chapters 5 and 6)
Green-house trials were carried out to test the hypotheses generated from field data. These trials considered both morphological and physiological adaptations of *L. uniflora* to algal loading.

Using the results of these two approaches, plus available information in the published literature, a model for the maintenance of *L. uniflora* populations in Scottish lochs was produced (Chapter 7).

Chapter 2:

The Study Sites

Chapter 2:

The Study Sites

Section 1
INTRODUCTION

This chapter describes the study sites in terms of published literature and abiotic conditions. A description of each of the lochs studied and a brief résumé of previous research is presented in Section 2. Section 3 describes the methodology used to measure abiotic parameters and the fourth section gives a brief description of the data collected over the two year period. These data are further considered in Chapter 4, where 2 years morphological field measurements of *Littorella* are discussed, in relation to measured environmental parameters in the four lochs.

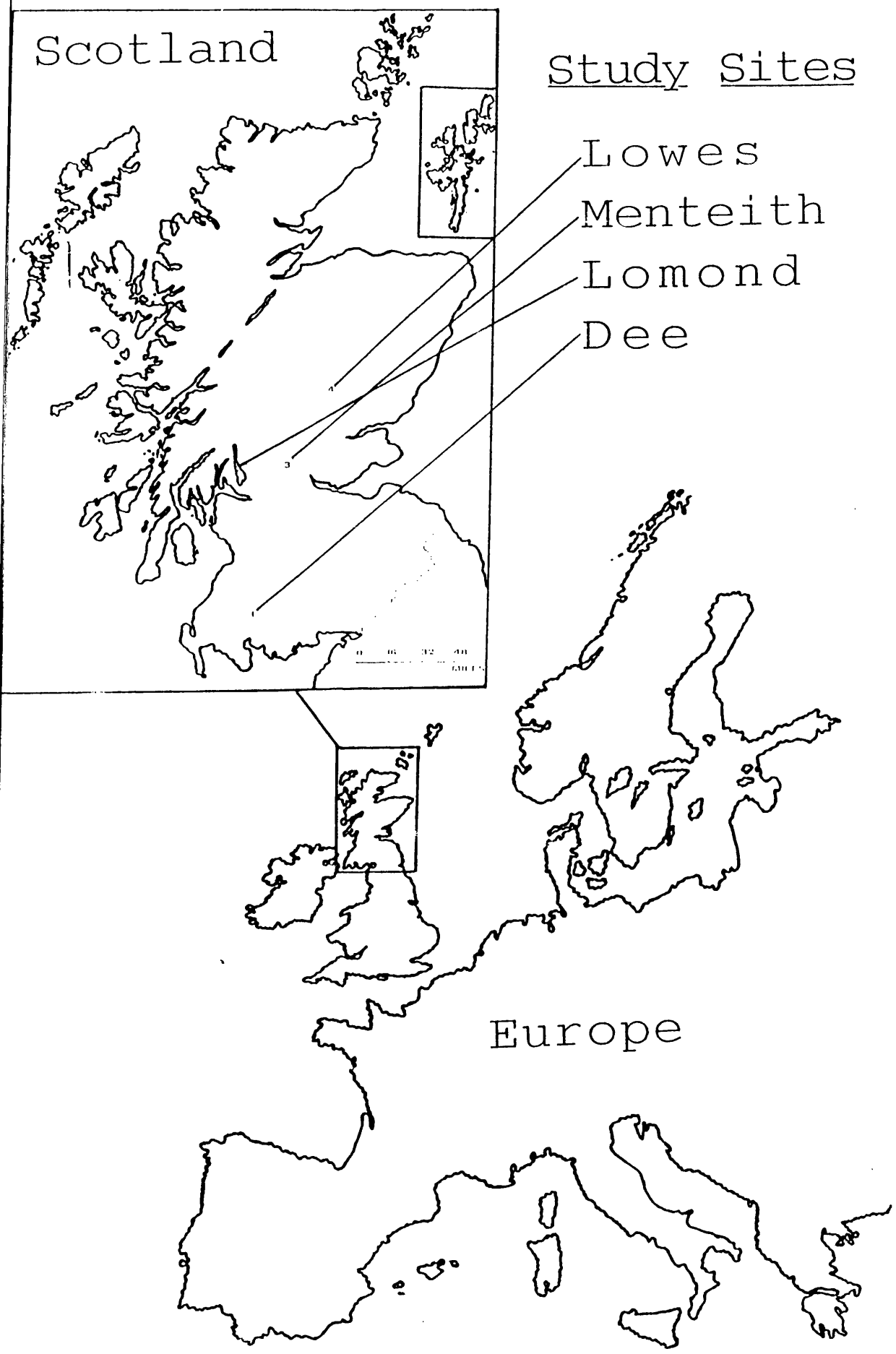
During the autumn of 1989, ten lochs were surveyed to determine their suitability for this study (See Appendix A). Four lochs were selected for work during 1990 and 1991. The main criteria used in the selection of sites were accessibility, the presence of a suitable macrophyte flora and trophic status.

The four lochs selected were as follow: Loch Dee (National Grid Reference (NGR) NX 470 970); the middle reaches of Loch Lomond (NGR NS 372 916); Lake of Menteith (NGR NN 572 007), and Loch of Lowes (NGR NO 046 440). The locations of the sites are shown in Figure 2.1.

Section 2
GENERAL DESCRIPTION OF THE STUDY SITES

2.1 **Loch Dee** lies in the mountains of Galloway in south east Scotland. Due to high annual rainfall levels (2232mm yr^{-1}), this loch is located in one of the areas in Scotland receiving the greatest loading of acid deposition (Loch Dee Project, 1989). The loch is moderately acidified and there has been much work carried out with respect to acidification and fisheries' management. This work, co-ordinated by the Solway River Purification Board (SRPB), has produced a mass of information including data on water chemistry (Lees 1991),

Figure 2.1 Location of the Study Sites


Scotland
Study Sites
Lowes
Menteith
Lomond
Dee
Europe

hydrology (Burns *et al.*, 1984), geology (Welsh & Burns, 1987; Marsden, 1990), aquatic macrophytes (Murphy *et al.*, 1986) and phytoplankton (Bailey-Watts & Kirika, 1991). The available data have been summarised by the Loch Dee Project Group (1993).

Interest was first raised in Loch Dee during the 1970s due to the decline of an excellent brown trout sport fishery. In 1979 the Loch Dee Project was set up to assess the effects of upland afforestation and acidification on surface water ecology and to develop the loch's potential as a brown trout fishery (Loch Dee Project, 1989).

The surrounding catchment area consists of basin and valley peats, with a depth range of 1 to 5m, which remain waterlogged throughout most of the year and are strongly leached. The underlying bedrock is predominantly granite and similar coarse-textured acid igneous rocks (Bown, Bibby & Shipley, 1982).

Sixty-seven percent of the total catchment lies above 304m. There are three main tributary burns; Dargall Lane, Green Burn (70% afforested) and White Laggan Burn (30% afforested). In non-afforested areas the vegetation is rough, low-quality, unimproved grazing (Bown, Shipley & Bibby, 1982). The water system is very poorly buffered with calcium levels around 2mg l^{-1}, so the pH of the water is largely dependent on that of the precipitation, resulting in large pH fluctuations in a matter of a few hours (Loch Dee Project, 1989; 1993).

In 1981 and 1983 powdered limestone was applied to the loch via the White Laggan Burn. In both cases an improvement in loch pH was initially observed but dropped to pre-liming levels within a period of two years. Future liming is unlikely due to the short-term effectiveness of the treatment caused by short retention time of the water in the system (Tervet and Harriman, 1988).

The loch basin is fairly shallow with a mean depth of 4.6m. In the west basin of the loch the maximum depth is over 10m, but the area in the east is mainly about 2m deep (Werrity & Maucotel, 1993).

The emergent macrophyte community fringing the loch is dominated by *Carex rostrata* Stokes, *Equisetum fluviatile* L. and *Ranunculus flammula* L. The

submerged aquatic macrophyte vegetation is dominated by *Juncus bulbosus* L. and plants of isoetid growth form, such as *Littorella uniflora* Ascherson; *Isoetes lacustris* L.; *Lobelia dortmanna* L. and to a lesser extent *Subularia aquatica* L. (Murphy *et al.*, 1986).

Phytoplankton studies during 1988 to 1990 showed the loch to have a low phytoplankton biomass with chlorophyll *a* levels rarely exceeding 4µg l^{-1}. This was attributed to high rainfall, low nutrient status and high flushing rate of the loch (see Table 2.1). The phytoplankton assemblages are dominated by small species of less than 2µm in diameter (picoplankton), which may be present in extremely high numbers (e.g. 100,000 ml^{-1}) (Bailey-Watts & Kirika, 1991; 1993).

2.2 **Loch Lomond** along with Lochs Awe, Ness, Morar & Shiel is one of the five largest lochs in Scotland (Maitland, 1981). It has the largest surface area and is the third deepest loch or lake in Britain. The loch was formed during the Pleistocene era (3 million years ago) by the erosive action of ice, and its catchment has a complex geology of sedimentary, metamorphic and igneous rock.

Loch Lomond is important to the surrounding area in terms of recreational facilities (Dickinson, 1991); as a water supply for several towns in the area (Mitchell, 1991); and as a source of electricity with the Sloy hydro-electric scheme having been in operation since the 1950s (Hamilton, 1988). The level of loch is controlled by a barrage across the River Leven south of Balloch (Mitchell, 1991).

Also of interest, is that Lomond is the only large loch to cross the Highland boundary fault, resulting in an interesting divide between the north and the south basins (Slack, 1957).

The north basin of the loch lies to the north of Inverbeg and Rowardennan and is 18km by 1.5km with a maximum depth of 180m at Tarbet. The steep sides of the loch in this region means there is only a small area of suitable depth for colonisation by aquatic macrophytes. Productivity by macrophytes in this region is consequently relatively low (Maitland, 1981).

To the south of Luss and Inchlonaig lies the southern basin which, in contrast to the northern basin, is wide (7km by 11km) and shallow (greatest depth 18m) and features many small islands. The surrounding catchment is of rich farming land, and treated sewage from eight small sewage plants is discharged directly into the loch (Hamilton, 1988).

The soil in the area of the study (between the islands of Inchtavannach and Inchmoan) is derived from slate, phyllites and other weak metamorphic rock. The islands are moderately rocky with brown forest soils, humic podzols and humic gleys in flushed hollows. Vegetation on the island is dominated by oak woodland and associated species with banks of *Myrica gale* L. at the water's edge.

The aquatic macrophyte flora is dominated by *L. uniflora* down to a depth of about 1.5m, with *L. dortmanna* and *J. bulbosus* also present in shallower water (down to about 0.5m). In deeper water, down to a depth of about 3.5m, depending on the height of the water table, the flora is dominated by *I. lacustris* interspersed with clumps of *Myriophyllum alterniflorum* D.C.. Throughout the macrophyte colonised area, below about 0.5m, large clumps of *Elodea canadensis* Michaux can be found colonising patches of sediment that have been exposed due to, for example, damage by boat hulls.

Phytoplankton studies carried out during 1971-72 (Maulood & Boney, 1979) showed the loch to be mainly dominated by diatom and desmid species. However, these authors recorded the appearance of *Anabaena circinalis*, a cyanobacterium, in the southern basin, which was attributed the possible onset of eutrophication. In the northern basin species (e.g. *Tabellaria flocculosa*) typical of oligotrophic water bodies were dominant.

2.3 **Lake of Menteith** is one of only two 'lakes' in Scotland and is probably most famed for its importance in outdoor curling circles. The lake lies 20km to the west of Stirling in a lowland plain of coarse clays and peat mosses drained by the River Forth.

On the whole the lake is fairly shallow with a mean depth of 6m, and a deep hole down to 23.5m to the north west of Inchmahome, the biggest island in the

lake. Because of this shallowness thermal stratification is not an important feature of the lake. The water remains isothermal and well oxygenated throughout the summer, except in the deep hole (Fozzard and Marsden, 1985; Maulood and Boney, 1981).

The lake supports a successful trout fishery and is an important tourism facility in the area, with holiday chalets and a hotel on the east shore. A site of important historical interest on Inchmahome also attracts many summer visitors with boat trips running frequently to the island during the summer months. The lake was designated a Site of Special Scientific Interest (SSSI) in 1991, and supports a diverse avian fauna.

The emergent vegetation in Lake of Menteith consists predominantly of *Phragmites australis* (Cav.) Trin ex Steudal, with stands of *Glyceria maxima* O. Holmb, *Schoenoplectus lacustris* (L.) Palla, *Typha latifolia* L. and *Carex rostrata* Stokes also present. *Schoenoplectus lacustris* ssp. *lacustris*, a locally rare plant, has also been recorded (Smith *et al.*, 1992). *Littorella uniflora* forms large swards in the shallow water with *Nitella translucens* (Persoon) Agardh, *Callitriche hermaphroditica* L., *Myriophyllum alterniflorum*, *Elodea canadensis*, *Sparganium natans* L., *Potamogeton alpinus* Balbis, *P. obtusifolius* Mert. & Koch and *P. perfoliatus* L. all commonly occurring in deeper water (NCC records). *Elatine hydropiper* L. has been recorded as part of the flora since 1981 (Newbold & Palmer, undated; Preston, 1987).

During the 1970s, complaints by local residents about cyanobacterial blooms and aegagrophilous *Cladophora* (cladophora balls) on the eastern shore, prompted the Forth River Purification Board (FRPB) to survey the water quality, phytoplankton, zooplankton and profundal benthos in 1981, 1983 and 1985. The conclusion of their report was that there had been no profound change in the physical and chemical condition of the loch since about 1970, although localised changes in areas of the lake may have occurred. The report classified the lake as mesotrophic to moderately eutrophic, with a tendency toward the former during winter and toward the latter during summer (Fozzard and Marsden, 1985). Routine surveys of water quality of the outlet of the eastern shore are still carried out on a monthly basis by the FRPB.

A study of the phytoplankton in 1972/1973 (Maulood & Boney, 1981) described the planktonic flora as primarily cyanobacteria to cyanobacteria/diatom in nature with spring and autumnal blooms of *Melosira*, *Asterionella* and *Fragilaria*, and three species of *Anabaena* producing large populations throughout the year.

2.4 **Loch of Lowes** is described as a fine example of a natural unpolluted mesotrophic loch (Harper, 1978; Nature Conservancy Council, 1985) and is located three miles north of the Highland Boundary Fault in Perthshire, near the village of Dunkeld. The loch is one of a group of three lochs (the others being Loch Craiglush and Loch of Butterstone) which have been designated SSSI due to the diversity of aquatic macrophytes. Part of the site is a Scottish Wildlife Trust Nature Reserve with excellent habitat for overwintering wildfowl. A pair of ospreys nested there until recent years (Nature Conservancy Council, 1985). In 1991 a new pair of ospreys successfully reared two chicks and returned in 1992 to breed again (A. Barclay, Scottish Wildlife Trust, pers. comm.).

The catchment geology consists mainly of Dalradian quartzes and mica-schists, with 50% heathland and upland grazing, 40% mainly coniferous woodland and 10% intensive agriculture. The highest ground in the catchment is 152m.

A survey of the submerged macrophytes carried out in 1973 (Harper, 1978) found *Littorella uniflora* to be present down to a depth of 2m and *Isoetes lacustris* to 3m. *Lobelia dortmanna* was present only at a depth of 1m. Species recorded in the depth range 0.5 to 5m were *Elodea canadensis*, *Myriophyllum alterniflorum* and *Potamogeton perfoliatus* L.. The deepest colonising macrophyte was *Nitella opaca* (Bruz.) Argadh, which along with *Potamogeton obtusifolius* was recorded in the 2.5 to 5m depth range. Sparse populations of *Callitriche hermaphroditica* L. were recorded between 4m and 5m depth.

A more detailed series of surveys carried out with the aid of snorkels in 1987/88 (James 1988) recorded all of the above species plus *Subularia aquatica*; *Potamogeton gramineus* L., *P. crispus* L. and *Eleocharis acicularis* (L.) Roemer & Schultes. This survey also revealed the presence of the rare annual *Najas flexilis* (Willd.) Rostkov & Schmidt of which there are only two other

records in Perthshire (Smith *et al.*, 1992). Both *Najas flexilis* and *Potamogeton filiformis* Pers. were recorded by Preston (1987).

Records from the 1970s (Harper, 1978) show the phytoplankton flora to be dominated by diatom genera, however more recent reports (Preston, 1987; James, 1988) record the presence of dense blooms of *Gleotrichia* sp., a cyanobacterium.

Table 2.1
Summary of the physical features of the four lochs

FEATURE	Dee	Lomond	Lowes	Menteith
length (km)	1.9	36	1.5	2.6
area (km^2)	1	71	0.9	2.6
shoreline (km)	7.1	154	4.4	9.2
maximum depth (m)	14.7	190	16	23.5
mean depth (m)	4.6	37	6.2	6
volume (Mm^3)	3.7	2628	5.4	15.9
catchment (km^2)	15.6	781	14.9	16.5
rainfall (mm/year)	2232	2200	1518	
mean retention time (year)	0.1	1.9	0.7	0.8
height above O.D.* (m)	225	9	100	16
latitude	55°10'N	56° 6'N	56°35'N	56°10'N
longitude	4°24'W	4°24'W	3°33'W	4°17'W

Sources of information:

Dee:	Loch Dee Project, 1989; Ordnance Survey, 1981
Lomond:	Maitland, 1981; Hamilton, 1988
Lowes:	Harper, 1978; Ordnance Survey, 1978b, 1983.
Menteith:	Ordnance Survey, 1978b, 1983; Fozzard & Marsden, 1985; Maulood & Boney, 1981.

*O.D. - Ordnance Datum

Section 3

SITE CHARACTERISTICS

3.1 Selection of Study Plots

Three fixed 10m x 10m plots were selected for study in each of the four lochs. Plots were marked by red painted wooden stakes on the shore. In order to alleviate the problems of changes in macrophyte physiology and morphology with depth (Søndergaard & Bonde, 1988; Spence *et al.*, 1973) these plots were located 1m below the shallowest depth of permanent colonisation of macrophytes such as *Lobelia dortmanna* (L.) and *Littorella* in the most sheltered region of the loch. The shallowest depth of colonisation of such macrophytes was deemed to be the best indicator of loch height and was determined at the most sheltered area of each loch to eliminate the effects of wave-induced water turbulence on colonisation (Spence, 1982).

It was impossible to select plots within each site that experienced the same wind exposure as this varied widely between each loch. The upland Loch Dee was generally the most exposed; wind exposure ratings of each of the plots are summarised in Table 2.2 at the end of this chapter (Weisner, 1987).

Study plots were selected on the basis of having a suitable population of *Littorella* colonising an area of 10m X 10m in region of 1m depth. It was decided that a suitable population was one where *Littorella* made up at least 60% of the macrophyte flora; although abundances of less than this were recorded occasionally in samples from Menteith plot 3 and all the Lowes plots. The locations of the plots within each of the study sites are shown in Figures 2.2a-d.

Figure 2.2
Location of Sites in the Four Lochs

Numbered clockwise from top right hand side
2.2a Loch Dee
2.2b Loch Lomond
2.2c Loch of Lowes
2.2d Lake of Menteith

Originally traced from Ordnance Survey 1:10,000 series maps

Dee:	1977a
Lomond:	1977b
Menteith:	1773, 1978a
Lowes:	1978b, 1983.

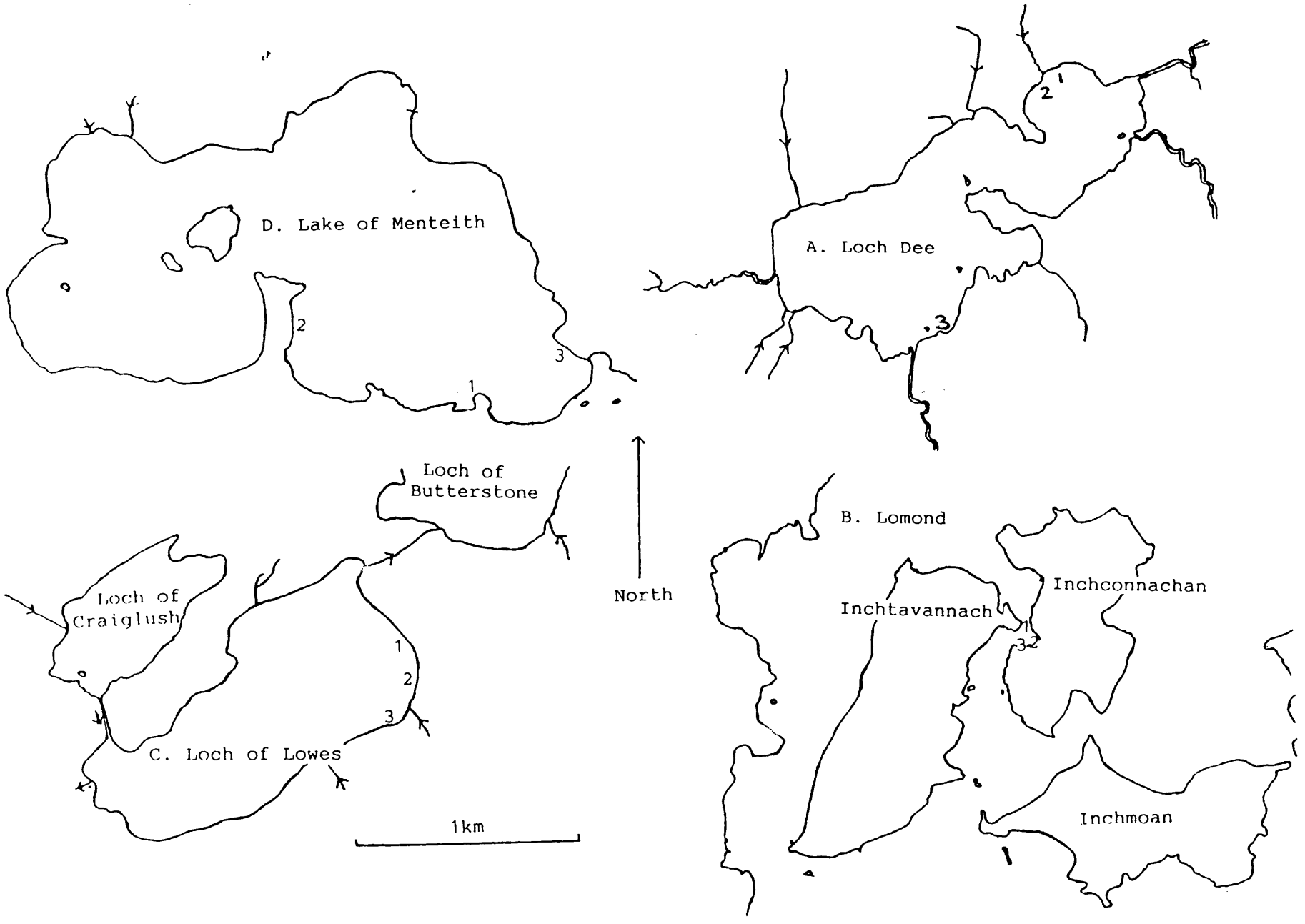

D. Lake of Menteith
2
3
1
A. Loch Dee
2
1
3
Loch of
Butterstone
Loch of
Craiglush
1
2
3
C. Loch of Lowes
North
1km
B. Lomond
Inchconnachan
Inchtavannach
1
3
2
Inchmoan
1

Figure 2.3
Loch Dee photographed from the Green Burn catchment in October 1990. Sites 1 and 2 are located on the opposite shore in the right hand basin.

Figure 2.4
Loch Lomond photographed from site 3 in August 1990. Site 1 is located on the right, with the Glasgow University Field Station catamaran on the left.

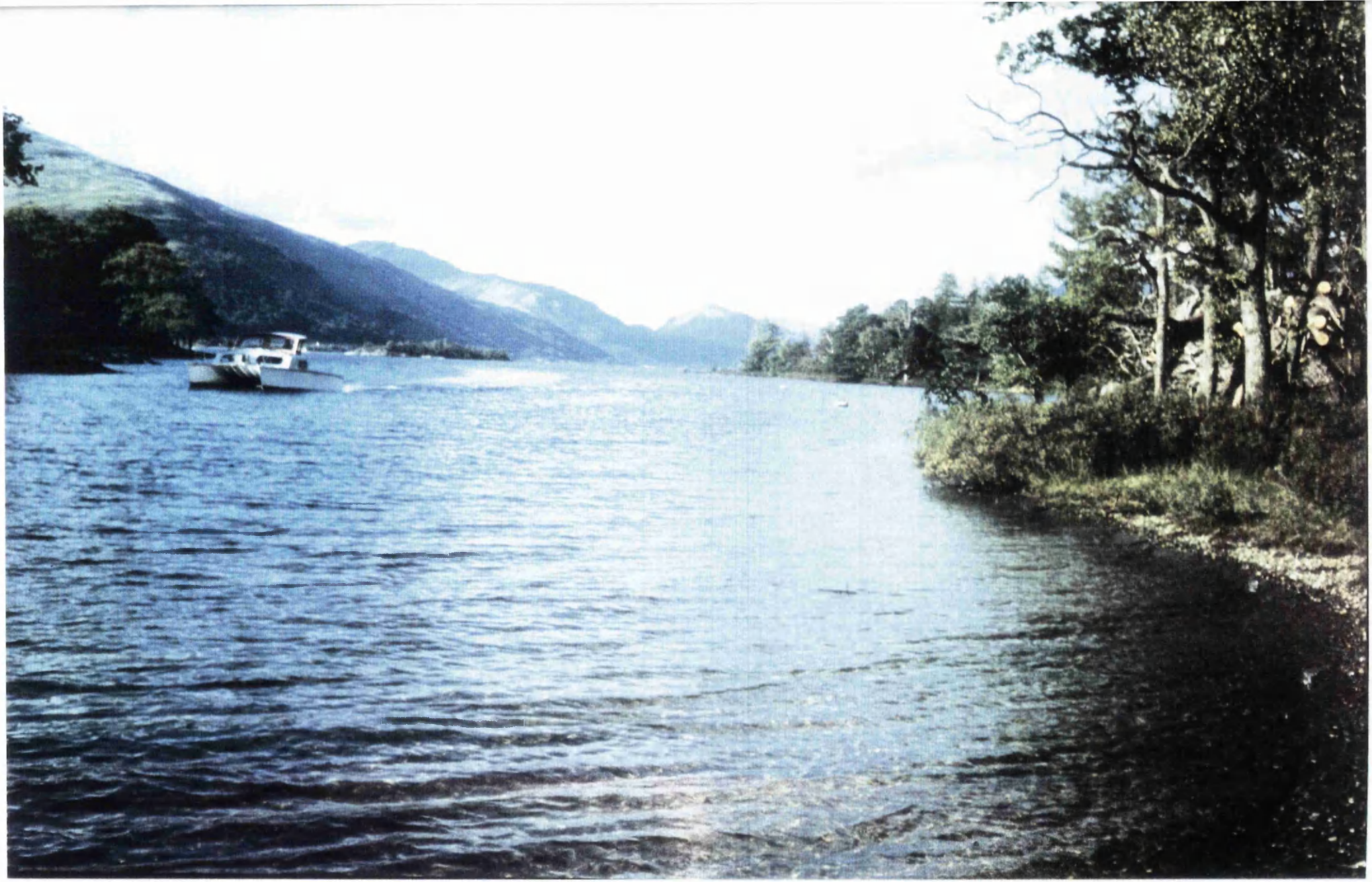
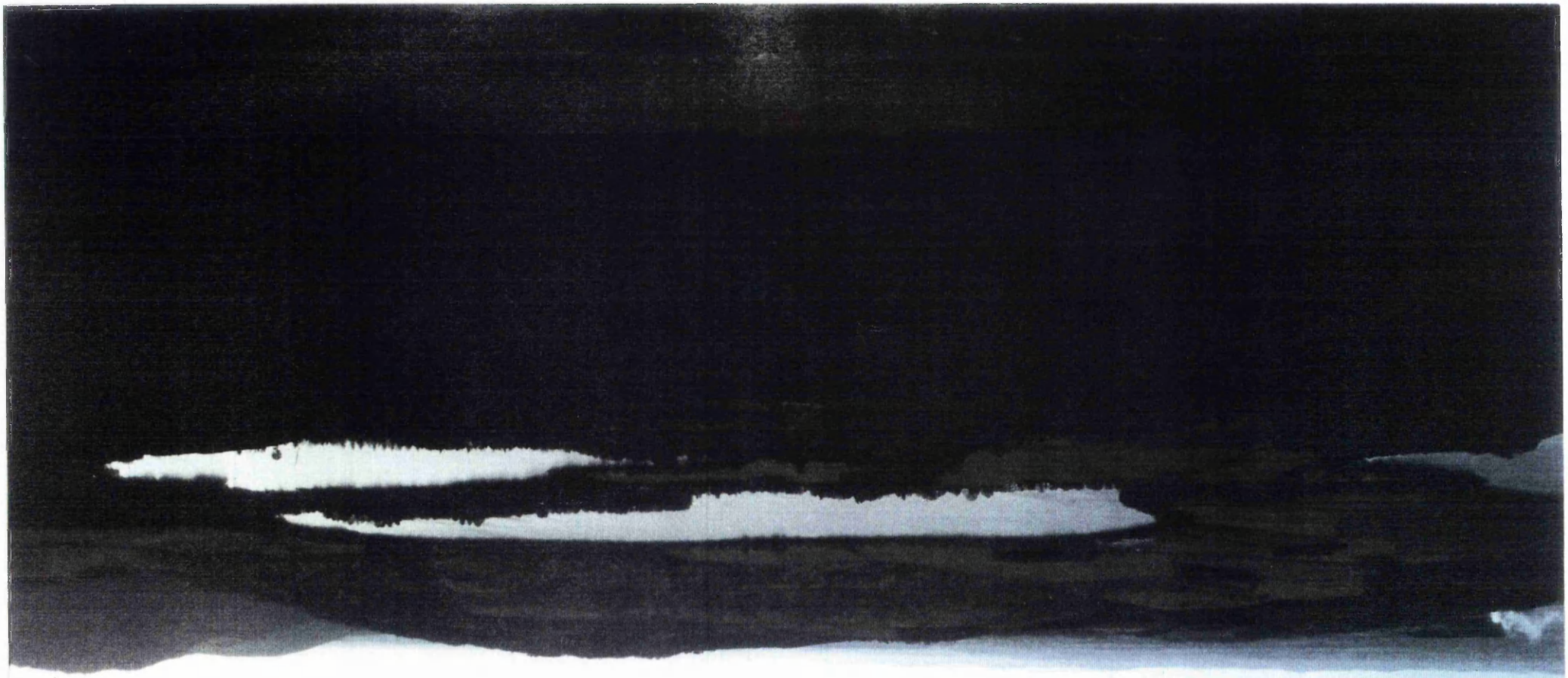


Figure 2.5
Lake of Menteith
Photograph taken from the A 873 road on the north shore, November 1992.

Figure 2.6
Loch of Lowes in the centre, with Loch Craiglush in the foreground and Loch Butterstone to the left. Photographed from Deuchary Hill, February 1992.



3.2 Sediment Characteristics

3.2.1 Methods

3.2.1a Sampling

Sediment samples were collected from the area surrounding the roots of macrophytes using a trowel and with the aid of SCUBA. Samples were placed in sealed polypropylene containers *in situ* and removed to the laboratory with the minimum of disturbance. The samples were then oven-dried at 105°C until a constant weight was achieved. Four replicates from each plot were sampled and at this stage equal weights of the four samples were pooled and stored in an airtight container until required for further analysis.

3.2.1b Organic Content - Loss on Ignition

This method follows that of Allen *et al.* (1986). After passing through a 2mm sieve, 10g aliquots of air-dried sediment samples, collected during August & September 1990, were placed in porcelain crucibles of known weights and dried overnight in an oven at 105°C. The samples were cooled in a desiccator and the oven dry weights were accurately determined using a Precisa 125A balance. Samples were placed in a muffle furnace at 600°C for 6 hours. The ignited soil was again allowed to cool in a desiccator and re-weighed. Two replicates of each sample were combusted.

3.2.1c Particle Size Analysis

The size of particles in the substrate of an area of lake depends on the degree of exposure of the site to turbulent mixing caused by wind/wave action and water currents. Once suspended in the water column, particles are sorted by a combination of wave action, turbulent mixing and long shore currents, with deposition dependent on the settling velocity. Settling velocity of a particle is size-dependent (see Allen, 1985).

Distribution of sediments in a lake can have profound effects on macrophyte colonisation and growth. Areas of fine sediments tend to have higher nutrient concentrations (Spence, 1982). In areas of severe exposure plants may be limited to species such as the isoetids, which are of small size (this helps reduce

mechanical damage) and with large root systems, which provide effective anchorage (Keddy, 1982).

The wind exposure rating at each site has already been considered (Section 3.1). However, as sediment distribution is influenced by more than wind-induced wave action, it is necessary to consider the sediment particle sizes that are present at each site as an indication of the sum of all the water movement mechanisms within the site.

Method for Particle Size Analysis

The method used was based on Kilmer & Alexander (1949). A known weight of oven dried sediment sample was passed through a 2mm sieve. The gravel fraction was weighed and from this the percent gravel could be calculated. The gravel was then discarded.

10g of <2mm air dry soil was placed in a preweighed 500ml conical flask and oven dried overnight at 105°C. An accurate sample weight was obtained after the sample had been cooled. Thirty percent (w/v) H_2O_2 was added in 10ml aliquots to dissolve any organic matter until no further reaction was observed; 30 minutes was allowed to pass between each treatment. The samples were then left overnight.

The sides of the beaker were washed with distilled water and the sample diluted to a volume of 40ml. The samples were then heated on a hotplate for 1 hour at 90°C ensuring the volume was maintained above 25ml by the addition of warm distilled water. After warming 20ml of 20% HCl was added to neutralise any remaining hydrogen peroxide.

The samples, which were now free of organic matter, were then placed on filter paper in a filter funnel and rinsed with hot distilled water until no chloride ions were present in the filtrate; this was determined by the addition of a few drops of $AgNO_3$ solution which forms a white precipitate in the presence of Cl^-. The samples were then washed into a 250ml conical flask, 20ml of 20% (w/v) sodium carbonate (Na_2CO_3) was added and the samples were boiled for half an hour and then shaken vigorously on a rotary shaker for 30 minutes to disperse any clumped particles.

After shaking, the soil samples were weighed and transferred to a 1000ml measuring cylinder and made up to a volume of 1000ml with distilled water. Each sample was shaken vigorously using a hand-plunger stirrer, then left to settle. After 4 minutes 48 seconds 25ml of the sediment suspension was removed from a depth of 10cm and placed in a 50ml beaker of known weight. After drying in an oven at 105°C this sample was weighed and this represents the silt and the clay particles present.

After a total of 4 hours had elapsed a further 25ml sample was removed from the measuring cylinder at a depth of 5cm and placed in a pre-weighed beaker, and then dried. This sample was weighed and represents the clay particles present.

After a further four hours the cylinder contents were siphoned off until 200ml remained. The contents of each cylinder were then transferred to a 1 litre beaker which was filled with water and the contents shaken. After 4 minutes 48 seconds the contents of the beaker were siphoned off until only 200ml remained - this was repeated until the water above the 200ml level became clear to the eye (between 10 and 30 washes).

The remaining sediment was then placed in a pre-weighed beaker and evaporated to dryness in an oven at 105°C. This fraction represents the fine and coarse sand. To obtain values for each, the sand fraction was passed through a 63µm mesh sieve after drying and the fine sand weighed.

The above proceedures were carried out and 20°C, and all settling times described in the above method are based on this temperature. Sedimentation times for other temperatures and particle sizes can be obtained from Tanner and Jackson (1947).

The following equations were used to calculate each component of the soil.

1. Clay % = Weight of clay x (1000/25) x (100/10)

2. Silt % = (Weight of clay + silt - Weight of clay) x (1000/25) x (100/10)

3. Fine sand % = Weight of fine sand x 100/10)

4. Coarse sand % = ((Weight of coarse sand + fine sand) - Weight of fine sand) x (100/10)

Sediment types were classified according to particle composition using the method described in the soil survey of England and Wales (Hodgson, 1974).

3.2.1d **Sedimentation Rate**

Sedimentation rate at each site was measured by placing three sediment traps in each of the of the study plots. A cylindrical trap of 10cm diameter and height 17cm (a one litre Azalon sample bottle with the top removed using a band-saw) was mounted on a 40cm x 40cm sheet of marine ply by three right angled stainless steel brackets. The trap was held in position by placing large stones around the edges of trap on the plywood. A photograph of the trap assembly is shown in Figure 2.7 (page36).

Traps were put in position during June 1990 and left in position until October 1992. The traps were emptied at the beginning and end of each sampling season with the exception of the 1991/1992 samples, which were left undisturbed for a full year.

In order to empty the traps, a SCUBA diver removed the weighting stones and gently raised the traps to the surface. Each trap was passed up to a person in a nearby boat where the contents were poured into a 1l sample bottle using a large funnel. The traps were then rinsed into the sample bottle with loch water and replaced by the diver.

The entire contents of the bottles were poured into pre-weighed 2l beakers and rinsed thoroughly to ensure all silt was transferred to the beaker. The samples were then left over-night to settle and excess water was siphoned off until 2cm remained above the sediment surface. Beakers were then placed in an oven at 105°C and evaporated to dryness. The beakers and sediment were weighed and results were expressed as grams sediment deposited per square metre per day (g m^{-2} day^{-1}).

3.3 Water Physico-chemistry

At each site visit the following water chemistry parameters were measured.

a) pH: measured using WPA Scientific Instruments CD62 pH meter.

b) Conductivity: measured using a Bibby SMC1 conductivity meter.

c) Photosynthetically Active Radiation (PAR): measured using a LI-COR model LI185B Quantum meter. Measurements were taken just below the water surface and at the plant level. Light measurements were taken at the plant level as opposed to deep water samples being taken in order to reflect changes in light attenuation due to phytoplankton blooms directly above the *Littorella* sample populations. The extinction coefficient (ξ) was calculated using the following equation:

$$\xi = \frac{\text{Log}_e(I_0/I_1)}{d}$$

Where: I_0 = sub-surface PAR
I_1 = PAR measured at depth
d = depth in metres I_1 measured at.

d) Surface water temperature: measured using a Gallenkamp Griffin mercury thermometer.

e) Dissolved oxygen: measured at plant level using a Bibby SMO1 dissolved oxygen meter in 1990 and a pHOX 62TE portable dissolved oxygen indicator in 1991.

f) Phosphates: Soluble phosphate was measured with an Auamerck phosphate test kit; range 0.25 - 3 mgl-1 soluble phosphate-phosphorus, accuracy +/- 0.125 mgl-1.

g) Nitrate concentration: measured using Merckoquant test strips; range 5-50 mgl-1 as nitrate-nitrogen, accuracy +/- 2.5 mgl-1 during 1991.

Figure 2.7

A sediment trap *in situ*. Photographed in Loch Lomond by Alan Bell, June 1991.

Section 4
RESULTS

Raw data for the survey results presented in this chapter are tabled in Appendix C and summarised in Table 2.2.

4.1 Sediment Characteristics

The exposure index as described by Weisner (1987) allowed a comparison of the relative exposure of plots to wind within each site. Plots with a high exposure index had a larger particle size and a lower sediment organic content than those with a low exposure index.

Loch Lomond contained the most sheltered sites, the highest sediment organic content, and the finest particle size. Sites 2 and 3 had an organic content of 10% and clay/loam or clay sediment types respectively. The sediment in Loch of Lowes was sandy with a sediment organic content of around 1%. Sites 1 and 2 in Loch Dee were both characterised as sandy/loam with a sediment organic content of 3.4 and 5.7% respectively. The more exposed site (3) had a loamy/sand sediment and a organic content of 0.5% which was 8-15% of that measured at the other two sites in this loch. Sediments in Lake of Menteith were sandy at sites 1 and 2, and loamy/sand in site 3. Sediment organic content in the three Menteith sites were 0.9, 4.0 and 2.1% respectively.

4.2 Sedimentation Rate

Loch of Lowes had a higher siltation rate than any of the other lochs. This site had a relatively sparse covering of macrophytes in comparison with the other sites, which had between 80 and 100% cover of macrophytic vegetation. Material contained in the collection vessel primarily consisted of sandy material that appeared to be resuspended sediment.

The higher sedimentaion rates at this site could be attributed to the resuspension of sediments that were not stabilised by rooted macrophytes. In particular, site 1, located next to the boat house, had a sedimentation rate of 2.5 g m^{-2} day^{-1}. This was more than double of that of any other recorded sedimentation rates in

the study lochs. The combined effect of activity round the boat house and sparse vegetation cover could account for these high levels of resuspended sediment.

Sedimentation rates in Loch Dee were lower than those at the other three sites, but were not significantly different from each other (one way analysis of variance $p > 0.05$) with all three sites having a siltation rate in the order of 0.24-0.30 g m^{-2} day^{-1}.

The siltation rates at sites 1 and 2 in Lake of Menteith were not significantly different from each other, however site 3 had a lower siltation rate. Cover, in this site (3) was 100% and plants were of the isoetid growth form. In contrast, the other two sites contained species such as *Myriophyllum alterniflorum* and *Potamogeton* spp. and contained areas of bare sediment (c. 20%).

The two sites located in the bay in Loch Lomond (sites 2 & 3) had a lower siltation rate than the site located in the channel between the two islands. The increased siltation rate in the site located in the channel can be attributed to boat traffic. Although a 5 mile per hour speed restriction has been imposed on this stretch of water, boats regularly travel through the channel at speeds well in excess of this.

4.3 Water Physico-chemistry

Phosphorous and nitrogen concentrations at no time exceeded the lower limits of the two test kits used (0.25 and 5mg l^{-1} P and N respectively) during the sampling period.

The four sites represent a gradient of water physico-chemistry, with Loch Dee at the lower end with a mean pH of 5.5 and conductivity of 40 µS cm^{-1}. Loch Lomond was intermediate with a maximum recorded pH of 7.3. Lake of Menteith and Loch of Lowes both had pH values greater than 8 during periods of high phytoplankton chlorophyll *a*. Loch Lomond, Lake of Menteith and Loch of Lowes all had a mean pH of about 7. Conductivity in the remaining Lochs - Lomond, Lake of Menteith and Lowes - were 50, 77 and 91µS cm^{-1}

respectively. Conductivity in all four lochs remained relatively constant throughout the sampling period.

Water temperature in Loch Dee was lower than the other three sites with recorded temperatures being 2-4°C lower than measured values at the other three sites. The lower temperature in Loch Dee reflects the high altitude of this site in comparison to the other three sites (see Table 2.1).

Table 2.2 **Summary of the abiotic characteristics of the 12 sites**

	SEDIMENT			
site	exposure rating	organic content (% dry weight)	sediment type	sedimentation rate ($g\ m^{-2}day^{-1}$)
Dee 1	6.1	3.36 (0.06)	sandy/loam	0.31 (0.54)
Dee 2	8.6	5.76 (0.24)	sandy/loam	0.44 (0.20)
Dee 3	18.7	0.54 (0.00)	loamy/sand	0.24 (0.04)
Lomond 1	16.1	8.14 (0.01)	sand	0.63 (0.11)
Lomond 2	3.6	10.18 (0.18)	clay/loam	0.22 (0.03)
Lomond 3	3.6	10.78 (0.00)	clay	0.38 (0.09)
Menteith 1	39.7	0.86 (0.01)	sand	0.56 (0.04)
Menteith 2	37.2	4.00 (0.06)	sand	0.41 (0.04)
Menteith 3	25.3	2.10 (0.06)	loamy/sand	0.14 (0.04)
Lowes 1	27.0	1.00 (0.02)	sand	2.46 (0.45)
Lowes 2	26.9	0.69 (0.01)	sand	0.72 (0.16)
Lowes 3	18.0	1.09 (0.03)	sand	0.92 (0.19)

	WATER			
site	pH	dissolved oxygen % saturation	conductivity $\mu S\ cm^{-1}$	water temp min- max °C
Dee1	5.6 (0.2)	93 (7.6)	41.7 (3.6)	6 - 15
Dee2	5.4 (0.1)	105 (6.3)	39.4 (2.7)	6 - 15
Dee3	5.5 (0.1)	109 (6.3)	40.3 (3.0)	6 - 16
Lomond 1	6.8 (0.2)	87 (7.4)	49.0 (2.7)	6 - 19
Lomond 2	6.6 (0.1)	89 (8.2)	50.5 (3.5)	6 - 19
Lomond 3	6.8 (0.1)	91 (6.9)	50.8 (3.6)	6 - 20
Menteith 1	7.2 (0.2)	93 (7.5)	77.5 (2.2)	7 - 19
Menteith 2	7.1 (0.2)	108 (7.1)	78.9 (3.8)	9 - 19*
Menteith 3	7.2 (0.2)	103 (9.2)	75.9 (2.8)	7 - 19
Lowes 1	7.2 (0.3)	88 (8.4)	90.5 (1.7)	8 - 18
Lowes 2	7.3 (0.3)	94 (6.6)	91.3 (1.9)	9 - 19
Lowes 3	7.4 (0.3)	93 (10.8)	91.2 (1.7)	8 - 18

Figures expressed as means(±se) for the two summer season sampling periods

Entire data set is tabled in Appendix C

* April data point missing

Chapter 3:

The Aquatic Flora of the Four Lochs

Chapter 3:

The Aquatic Flora of the Four Lochs

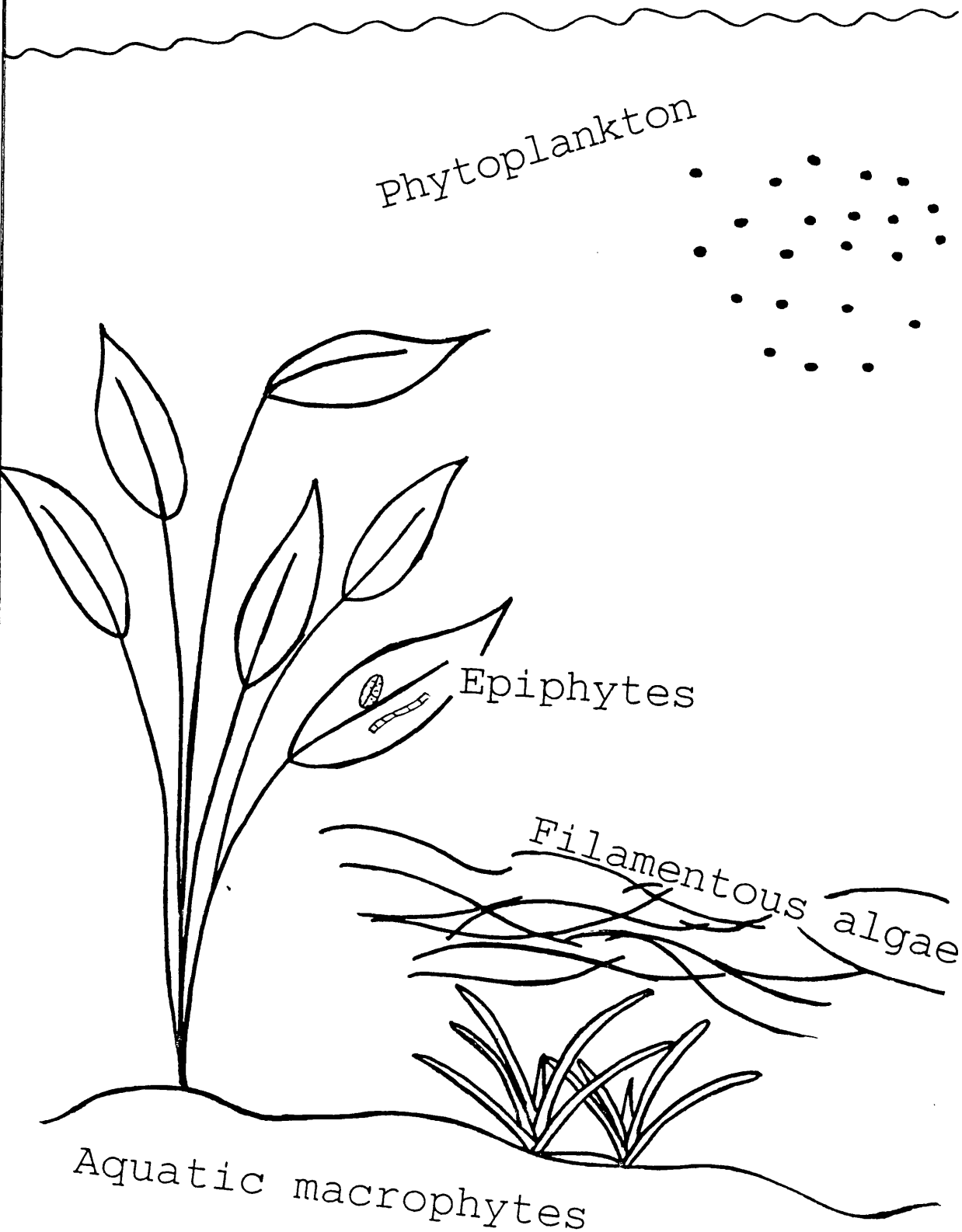
Section 1
INTRODUCTION

Four components of the aquatic flora were considered:

1. **Macrophytes**: these consist of large plants readily visible to the naked eye and for the purposes of this study included charophytes and all vascular plants. These were studied throughout the 1990/91 growing seasons.
2. **Phytoplankton**: this component is usually microscopic, although some colonies may be distinguishable to the naked eye. The phytoplankton are free-floating in the water column and their movement is largely controlled by wind and water currents (Blackmore & Tootill, 1984). Phytoplankton chlorophyll *a* was monitored during the summers of 1990/91.
3. **Epiphytes**: epiphytes are plants that grow attached to the surface of other plants. In the freshwater environment these plants are predominantly microscopic algae. Percentage cover estimates were carried out using a binocular microscope and verified by observations of Scanning Electron Micrographs of samples taken in July 1991.
4. **Filamentous algae**: under certain conditions large swards of free floating filamentous algae (metaphyton) form and grow over the surface of the plants. During 1990 filamentous algal growth was observed in two sites - Loch Dee and Loch of Lowès. During 1991 this was quantified in terms of dry weight per m^2.

Figure 3.1 gives a summary of the spatial distribution of each of these components within a water body.

Figure 3.1 The Spatial Distribution of the Flora



Section 2

METHODS

2.1 Sampling Regime

Sampling was carried out from the beginning of April to the end of October in 1990, and from April to mid September 1991. Sites were visited once every six weeks in 1990 and once every five weeks in 1991.

2.2 Macrophytes

2.2a Surveys

Each site was surveyed in 1989 (see Appendix A) by grapnel hauls, and again during November 1990. For the 1990 survey two divers surveyed the target area for at least 30 minutes. Plants were recorded on a presence/absence basis and the maximum depth of colonisation was recorded using a US Divers dive timer, which records depth to an accuracy of 0.1m. Measured depth was adjusted to that below the predicted zero depth based on the criteria described in Chapter 2 section 3.1. During the summer of 1990 species samples were collected for identification. Sheldon & Boylen (1978) found SCUBA diving to be almost twice as efficient for species diversity estimates when compared with random quadrats.

2.2b Biomass & Species Composition Along the 1m Isobath.

Biomass samples were collected from a depth of 1m from each of the plots at each site visit (4 each in 1990 and 1991). Initial samples were collected from the boat using an Ekman grab (see Blanquist, 1990; Ekman, 1911). However, SCUBA observations showed the grab to be effective in soft sediments only. Subsequently, four 25cm x 25cm (area = 0.0625 m^2) biomass samples were collected using a trowel and with the aid of SCUBA.

In 1990 species were recorded at each site then sorted into *Littorella* and 'others'. Plant material was washed in tap water to remove any sediment and dried in a Griffith-Grundy oven at 90°C until a constant weight was obtained (usually about 5 days). The % abundance of *Littorella* in terms of dry weight, and the standing crop

in each plot were calculated. During 1991 a similar protocol was followed except that all plant species as well as *Littorella* were weighed individually, so the species composition of the samples could be determined on a dry weight basis.

Further treatments and analysis of *Littorella* samples are described in Chapter 4.

2.3 Phytoplankton

There are several methods available for the study of phytoplankton biomass: direct counts: chlorophyll *a* determination: estimates of biovolume: measurements of ATP levels. Each method will produce different results for the same lake sample (Aleya *et al.*, 1988), and the technique employed for phytoplankton studies depends entirely on the aim of the study (Billington, 1991).

In this study experiments were conducted to determine if phytoplankton shading in the water column affected the growth of aquatic macrophytes. Chlorophyll *a* is the major pigment in freshwater phytoplankton. Attempts have been made to relate chlorophyll *a* concentration of phytoplankton to the attenuation of PAR in a water column. However, variable results have been obtained due to the importance of other factors such as cell size and the proportion of cell pigments composed of chlorophyll *a*. For example, cells such as diatoms contain high concentrations of the carotenoid fucoxanthin, so the importance of chlorophyll *a* as a light harvesting pigment is less than in green algae where levels of chlorophyll *a* are proportionally higher (Kirk 1975a, 1983).

Chlorophyll *a* levels were measured throughout both growing seasons, as an indicator of phytoplankton density.

2.3a Chlorophyll *a* Determination

2.3a i Collection and Filtration

Two surface water samples were collected from each plot at each site visit using 5l opaque plastic bottles. Opaque plastic was used in order to prevent light penetrating to the sample, to reduce the likelihood of degradation of chlorophyll to

phaeophytin. Samples were stored at 5°C in dim light until filtration could be carried out (within 6 to 18 hours).

5l of sample were filtered through a 'Millipore' filter apparatus connected to a Speedivac Vacuum Pump, and fitted with glass-fibre filter paper (Whatman GF/C). Glass fibre filter paper was selected as it has a high flow rate and is resistant to clogging (Brock, 1983). Lenz & Fritsche (1980) found glass fibre filter papers to have a lower retention of phytoplankton (10%) than 0.45µm membrane filters, however Holm-Hansen & Reiman (1978) found there to be no difference between the two filter types, and Hilmer and Bates (1989) found recovery rates from glass fibre filter papers to be the best of all the filters tested. A few drops of magnesium carbonate were added to the sample prior to filtration to prevent chlorophyll degradation due to low pH.

Samples were stored in a freezer (-20°C) until chlorophyll extraction could be carried out. This had the two-fold function of preserving the samples for up to six months (Lenz & Fritsche, 1980) and breaking cell walls to aid chlorophyll extraction (Reiman & Ernst, 1982). Holm-Hansen & Reiman (1978) found there to be no losses of chlorophyll in frozen wet samples. Chlorophyll was extracted from all samples within six months of collection.

2.3a ii **Extraction**

The extraction method was based on Hipkins & Baker (1986). Methanol was selected as the extraction solvent as this has a high efficiency of extraction compared with either ethanol or acetone (Holm-Hansen & Reiman, 1978; Rieman, 1980; Marker & Jinks, 1982) and chlorophyll *a* remains stable in methanol for up to 24 hours (Rieman, 1980). Chlorophyll *a* estimates were uncorrected for phaeophytin as this can lead to increases in estimation errors due to possible negative phaeophytin values (Bührer, 1991).

Filter papers were cut into approximately 5mm x 5mm pieces using a sharp new razor blade, placed in aluminium foil-covered test tubes containing about 5ml methanol (the volume of methanol was varied according to the density of algal sample), stoppered, then placed in a water bath at 70°C for twenty minutes. The

methanol/chlorophyll solution was then transferred to a clean, foil-wrapped, stoppered test tube. A second extraction was then carried out on the filters and the resultant solution from this added to the first extract.

The extracts were then centrifuged at 3,000g for five minutes to ensure there was no debris in the solution that could interfere with spectrophotometer readings. The supernatant was then transferred to a volumetric flask and made up to a known volume (usually 5 or 10ml).

Absorbance at 650nm and 665nm was measured using a Shimadzu UV-160A UV-visible recording dual beam spectrophotometer (bandwidth 2nm) in matched glass cuvettes with a methanol blank. Chlorophyll *a* concentration was calculated using the equation as described in Hipkins & Baker (1986).

µg Chl *a* per ml methanol = (16.5 * Abs665) - (8.3 * Abs650)

The lowest limit of accuracy for this method was 2µgl-1 loch water. Results are expressed per litre loch water.

2.4 **Epiphytes**

2.4a **Surface Cover Estimates**

Five intact *Littorella* plants were collected from each site with the aid of SCUBA and placed in a polypropylene container *in situ* during July - September, 1991.

Intact leaf surfaces were examined using a Nikon binocular microscope, percentage cover of epiphytes was estimated and expressed using the Braun-Blanquet scale (see Moore & Chapman, 1986).

2.4b Scanning Electron Microscopy

In order to verify epiphyte percentage cover estimates on leaves of *Littorella*, Scanning Electron Micrographs (SEM) were taken of the second-youngest leaves of one plant from each area within each study site (a total of 12 samples) during July 1991. Preparation of stubs was carried out by Mr E. Robertson of the Electron Microscopy Unit at Glasgow University, using a standard protocol as follows:

Specimens for SEM were fixed in 3% gluteraldehyde in 0.2M phosphate buffer. After 12-16 hours specimens were rinsed three times for a period of ten minutes each in the same buffer, and post fixed in 2% osmium tetroxide in water for three hours. The specimens were rinsed in distilled water then stained in 0.5% uranyl acetate for 1-2 hours. After staining specimens were dehydrated through an acetone series and critical point dried.

Specimens were fixed to SEM stubs by double sided 'Sellotape' and gold coated in a sputter coater for 8 minutes (approximate thickness of gold: 60-80nm). The specimens were then viewed in a Philips 500 SEM at an accelerating voltage of 3 or 6kV.

2.5 Filamentous Algae

During 1990, the development of the filamentous algal mats were observed, but not quantified. During 1991, filamentous algal biomass was sampled using a 17cm diameter polypropylene collar which was lowered very gently over the plant material to avoid disturbing the algal mat. The algal mat was subsequently removed using a small sieve and placed in a 'self-seal' polythene bag. Three replicates per plot were sampled.

Several collar sizes were tested. Small diameters were difficult to use as the width was to small to allow comfortable use of the small sieve. Larger diameters contained too much material to be removed without excessive losses occurring as a result of algae floating away. Sampling difficulties due to algae entanglment in *Littorella* leaves resulted in an underestimation of algal biomass by about 30%.

Measurement of light attenuation by filamentous algae was attempted in the field but proved to be unreliable due to algal disturbance after positioning the sensor. In order to measure light attenuation in the laboratory, filamentous algal samples were resuspended in the same area as that from which they were collected. This also proved unsatisfactory, as after collection the algae tended to form clumps. It was concluded that accurate measurements could not be made and so no estimates of light attenuation by filamentous algae were carried out.

Filamentous algae samples were collected during August/September 1991, when the filamentous algal biomass was maximal. The dominant species in the samples were identified in the laboratory with the aid of a key (Prescott, 1970).

2.6 *Cladophora* Balls

As mentioned in Chapter 2, aegagropilous *Cladophora* (*Cladophora* balls) are present in Lake of Menteith. These structures were present to a great extent in water deeper than 1m, although small amounts were present in the site plots. During windy weather large volumes of these balls are washed onto the shoreline. The main aim of this study was to investigate the effect of shading by algae on submerged macrophytes, and *Cladophora* balls which in this system did not appear to shade macrophytes were not sampled.

2.7 Data Analysis

With large data sets the use of standard statistical techniques that only consider one variable at a time is tedious and ineffectual. Multivariate statistical techniques allow many variables to be considered together and thereby improve the efficiency and senstivity of the analysis (Gauch, 1986).

Detrended Correspondence Analysis (DCA) developed by Hill & Gauch (1980) is an indirect ordination technique appropriate to the analysis of species data in several sites which overcomes some of the problems caused by mathematical artefacts in earlier ordination techniques such as Correspondence Analysis (ter Braak, 1987).

DCA was carried out on the presence/absence data obtained in 1990 and on the percentage abundance data collected in 1991 using the CANOCO package (ter Braak, 1988). The effect of seasonality was taken into consideration by treating the four sampling periods as blocks, and effects due to these were consequently removed from the analysis using the block effect option of the programme.

Section 3:
RESULTS

3.1 Macrophytes

3.1a Maximum Depth of Colonisation

Loch Lomond was the most deeply colonised with macrophytes extending down to a depth of 4m. Poor weather conditions at the time of sampling at Loch Dee prevented the maximum depth being determined, however macrophytic vegetation was observed in the west basin at a depth in excess of 2.8m. Maximum colonisation depths in Lake of Menteith and Loch of Lowes were 2.7m and 3.2m respectively.

3.1b Interpretation of Detrended Correspondence Analysis

As the sampled area at each site was restricted to the 1m isobath discussion of results obtained from analysis of this data cannot be extrapolated to each loch as a whole. As this study was primarily concerned with *L. uniflora*, the samples collected were biased and consequently, are not representative of the whole loch community. The absence of a species from one sampling period to the next is likely to be a reflection of the sampling procedure, rather than representing real changes in the aquatic flora. Despite these limitations to the data set, the DCA plots do highlight some interesting features.

Figure 3.2 depicts the DCA plot of species and sites sampled in 1990. *Littorella uniflora* and *Isoetes lacustris* lie in the centre of the plot as *L. uniflora* is present in all the sampled sites and *I. lacustris* is present in all but 4 of the sites.

Legend for Figures 3.2 and 3.3

X Loch Dee

■ Loch Lomond

☒ Lochs Dee and Lomond at the same point

Δ Lake of Menteith

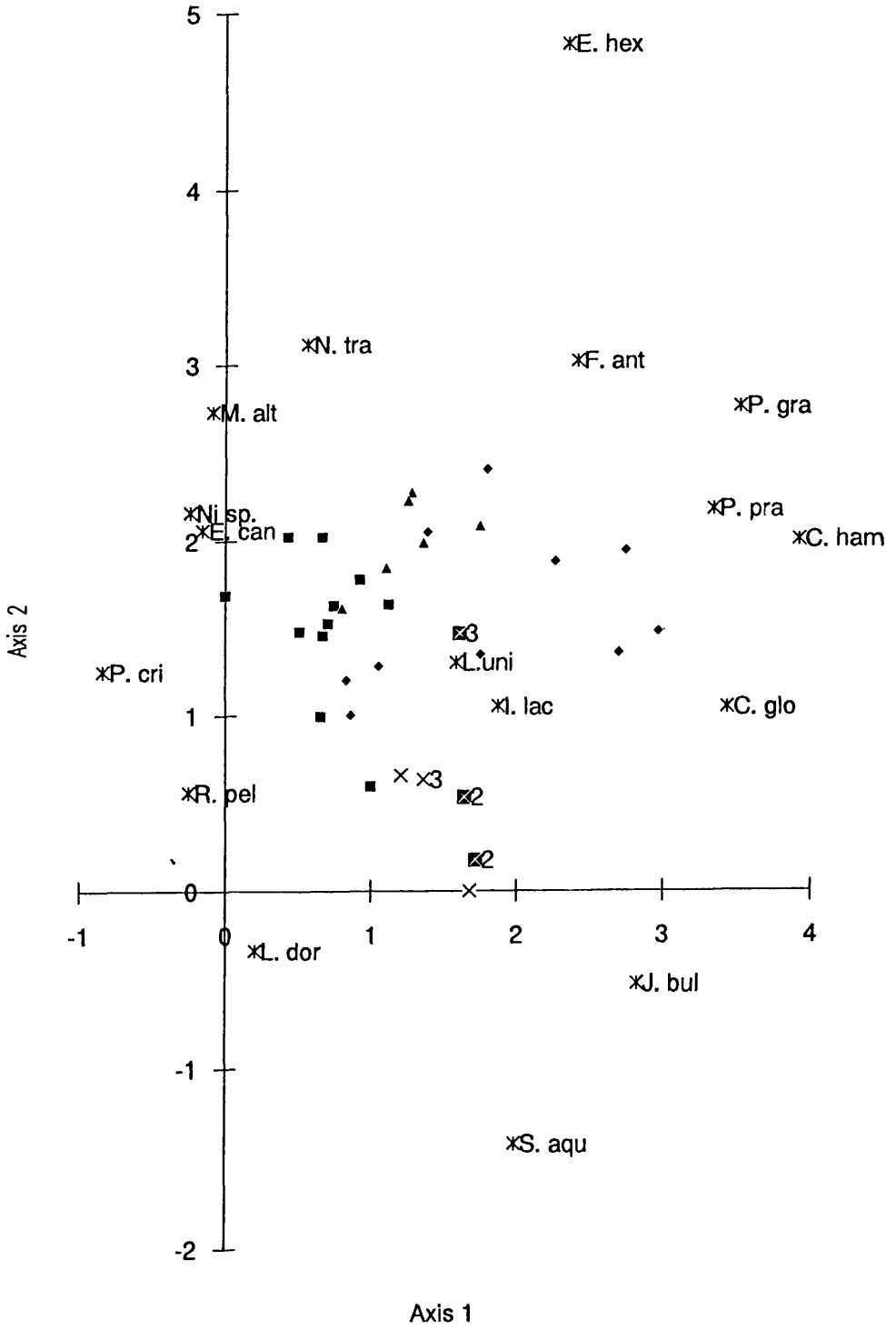
◆ Loch of Lowes

Where two points coincide the number of duplicated points is represented by a symbol is written to the right hand side of the relevant point.

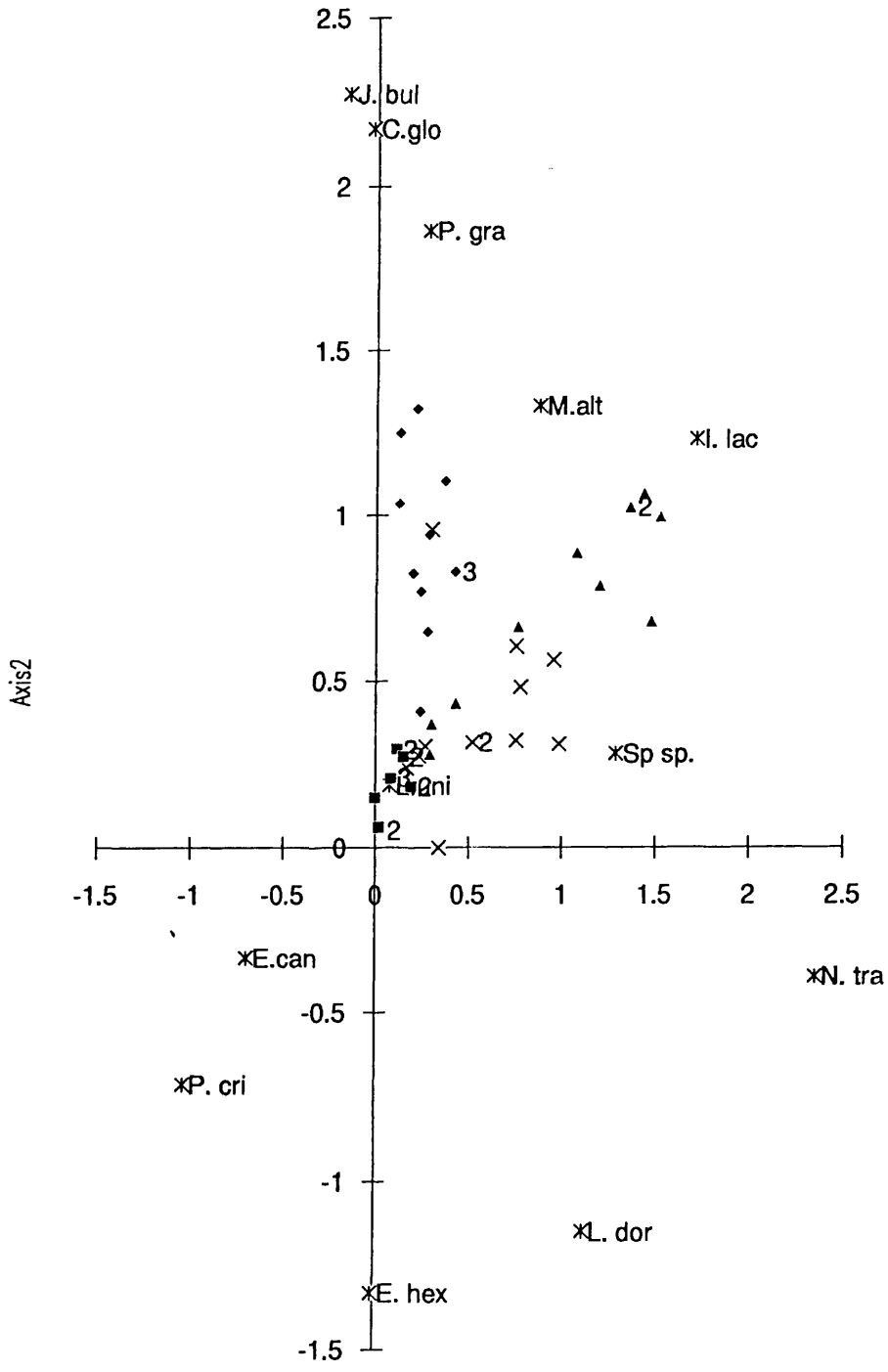
Key to species in Figures 3.2 and 3.3

C. ham	*Callitriche hamulata*
C. glo	*Chara globularis*
E. hex	*Elatine hexandra*
E. can	*Elodea canadensis*
I. lac	*Isoetes lacustris*
J. bul	*Juncus bulbosus*
L. uni	*Littorella uniflora*
L. dor	*Lobelia dortmanna*
M. alt	*Myriophyllum alterniflorum*
N. tra	*Nitella translucens*
P. cri	*Potamogeton crispus*
P. pra	*Potamogeton praelongus*
R. pel	*Ranunculus peltatus*
Sp. sp	*Sphagnum sp.*
S. aqu	*Subularia aquatica*

Figure 3.2 DCA of Species Along 1m Isobath 1990



DCA 1991


2.5
2
1.5
1
0.5
0
-0.5
-1
-1.5
-1.5
-1
-0.5
0
0.5
1
1.5
2
2.5
Axis2
Axis 1
J. bul
C.glo
P. gra
M.alt
I. lac
Sp sp.
E.can
N. tra
P. cri
L. dor
E. hex

Although the four lochs make up four discernible groups, there is some overlap between each group. The length of the primary axis is 2.97 standard deviations. Sites that differ by 4 standard deviations will have no species in common and represent different community types (ter Braak, 1986; 1987). From this it is apparent that the four lochs represent one basic community type with species differences in each of the lochs.

The Loch Dee sites are positioned towards the middle of the plot just below the centroids for *I. lacustris* and *L. uniflora* indicating the presence of *Lobelia dortmanna* and *Juncus bulbosus* in the majority of the samples.

Loch Lomond sites are positioned to the left, their location being influenced by the presence of species such as *Myriophyllum alterniflorum*, *L. dortmanna* and *Elodea canadensis*.

Loch of Lowes sites form an elongated group parallel to the first axis with sites containing *Potamogeton gramineus*, *Potamogeton praelongus* and *Callitriche hamulata* being located to the right of the group.

Lake of Menteith sites were located in the middle of the biplot and overlapped with both the Dee and Lowes site centroids. The most commonly occurring species in this loch during 1990 were *L. uniflora*, *I. lacustris*, *L. dortmanna* and *Nitella translucens*.

The first axis of the DCA for 1991 has an eigenvalue of 0.3841, indicating a poorer separation of species than in the 1990 DCA (eigenvalue of first axis = 0.4798) however the grouping of the sites on the biplot shows a clearer grouping between the lochs, with fewer sites overlapping. The use of percentage abundance data, as opposed to presence/absence data, has allowed a clearer differentiation between the four lochs.

Again *L. uniflora* is located in the centre of the plot, in this case *I. lacustris* was positioned at the edge of the plot, as this species did not occur in so many of the sites sampled in 1991 compared with 1990. *Potamogeton perfoliatus* and

Subularia aquatica are both rare species in the data set and have been omitted from Figure 3.3 in order to expand the centre of the biplot for clarity. Rare species have little influence on the overall analysis (ter Braak, 1987).

Centroids for Loch Lomond sites are positioned in a tight group around the *L. uniflora* centroid. Percentage abundance plots (Appendix D2biv-vi) for Loch Lomond show the area sampled in this loch to be poor in species with no less than 90% of the biomass consisting of *L. uniflora.* Figure 3.8 shows the uniform sward of *Littorella* at Site 1 in Loch Lomond.

Loch Dee sites are located closer to the *I. lacustris* centroid than the Lomond sites indicating a higher percentage abundance of this species in these sites (see Appendix D2bi-iii). *L. dortmanna* is also a common in Loch Dee.

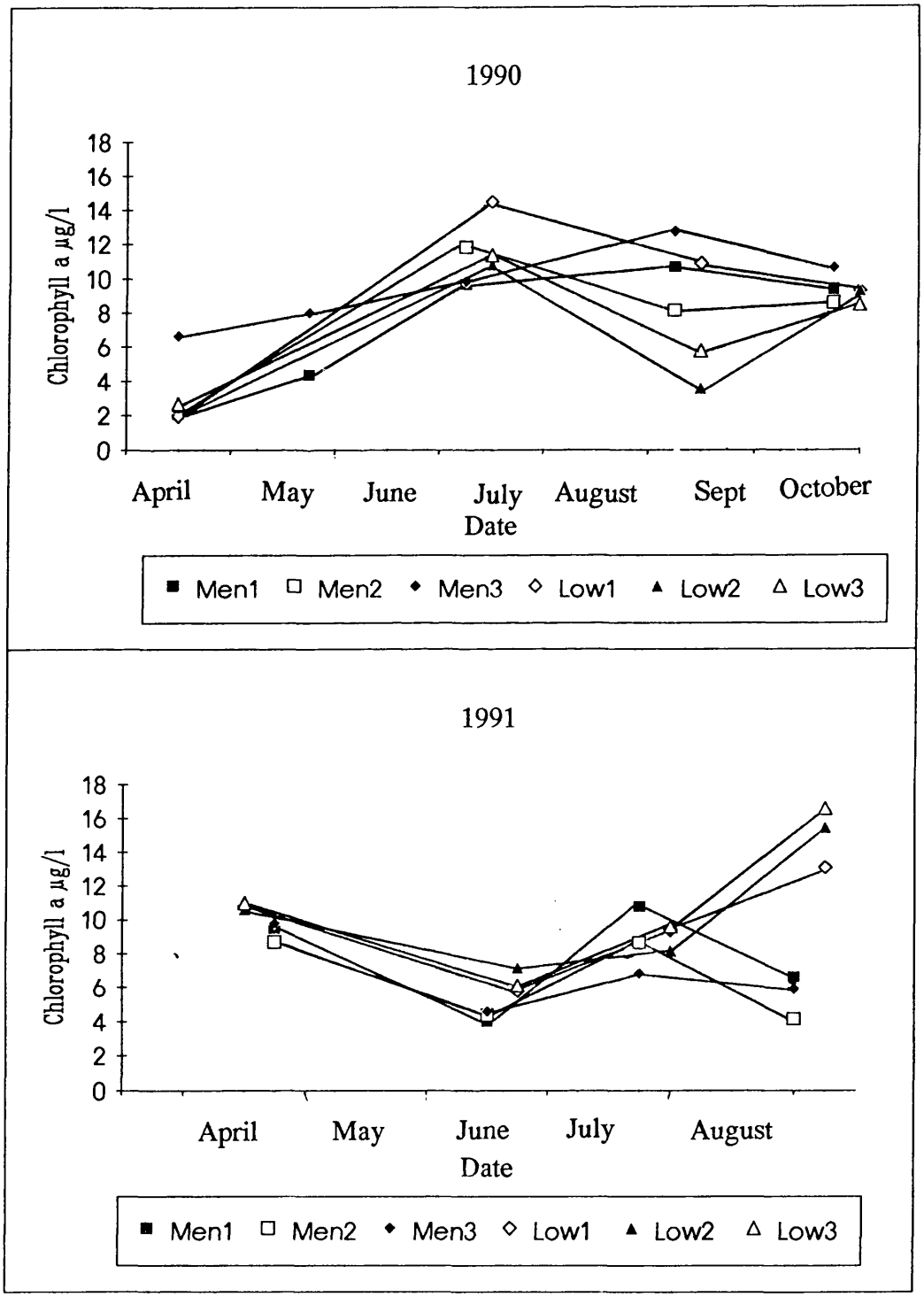
Lake of Menteith sites in 1991 were the only where display any pattern emerged in their distribution relative to the sampling plots. Plots 1 and 3 were located closer to the *I. lacustris* centroid than plot 2 indicating the higher percentage abundance of this species in these two plots (see Appendix D2bvii-ix).

Loch of Lowes sites are positioned higher on the second axis, but lower on the first axis, largely due to the influence of *J. bulbosus*, *Chara globularis* and *P. gramineus.* Loch of Lowes had the highest species diversity along the 1m isobath of the four lochs studied, with 5 species recorded at each site visit and 9 species recorded in July 1991 at Plot 1 (Appendix D2bix-xii).

3.2 **Phytoplankton**

Chlorophyll *a* levels in Loch Lomond and Loch Dee only exceeded 2µgl^{-1} during the month of June in both 1990 and 1991. In June 1991 an apparent peak of about 10µg Chl *a* l^{-1} was observed in Loch Dee, however this was caused by a gale force wind mixing the filamentous algal mat into the water column at plots 1 & 2. This increase was not observed at site 3 where algal mat formation was not extensive. At all other times sampled, the levels of chlorophyll *a* were lower than the limit of detection of the extraction method.

Figure 3.4 Phytoplankton Chlorophyll a
Loch of Lowes & Lake of Menteith 1990 and 1991



Standard error bars omitted for clarity

Loch of Lowes and Lake of Menteith both showed large variations in phytoplankton chlorophyll *a* (Figure 3.4). In Loch of Lowes during 1990 the bloom began to develop in April, then reached a peak in late June/early July, declined in late August, then had a small secondary peak in October. In the first half of the sampling season Menteith followed a similar pattern of phytoplankton development as Loch of Lowes, but after June chlorophyll *a* levels remained high until October.

In 1991 both Lake of Menteith and Loch of Lowes bloomed early with chlorophyll *a* levels around 10μgl^{-1} in April. Chlorophyll *a* levels in both lochs decreased during June to rise again in July. After July the chlorophyll *a* levels in Lake of Menteith began to decline, whereas those in Loch of Lowes continued to rise to a peak of approximately 15μgl^{-1} in September. Analysis of variance showed no significant difference between Lowes and Menteith chlorophyll *a* levels during the first three sampling visits of 1991 ($P > 0.05$).

3.3 Epiphytes

The use of the binocular microscope provided a means of estimating the epiphytic colonisation of *Littorella*. Figures 3.5 and 3.6 show scanning electron micrographs of *Littorella* leaf surfaces that scored 1 and 5 on the Braun-Blanquet scale respectively.

Plants from Loch of Lowes had the most extensive epiphyte covering, with a score of 5 being obtained for all plants observed. Lake of Menteith sites were also heavily colonised, although site 3 generally had a lower score of 4. Plants from all sites in Loch Lomond were sparsely colonised and scored 2. Loch Dee sites 1 and 2 scored 3, and the third site located in the more wave washed area of the loch had very low colonisation and scored 1. No increases in epiphyte cover were observed from June to September 1991 on the leaves of *Littorella* in these four sites.

Figure 3.5 Scanning Electron Micrograph of *Littorella* Surface Scoring 1 on the Braun-Blanquette Scale. Scale-bar: 1 division = 10 μm.

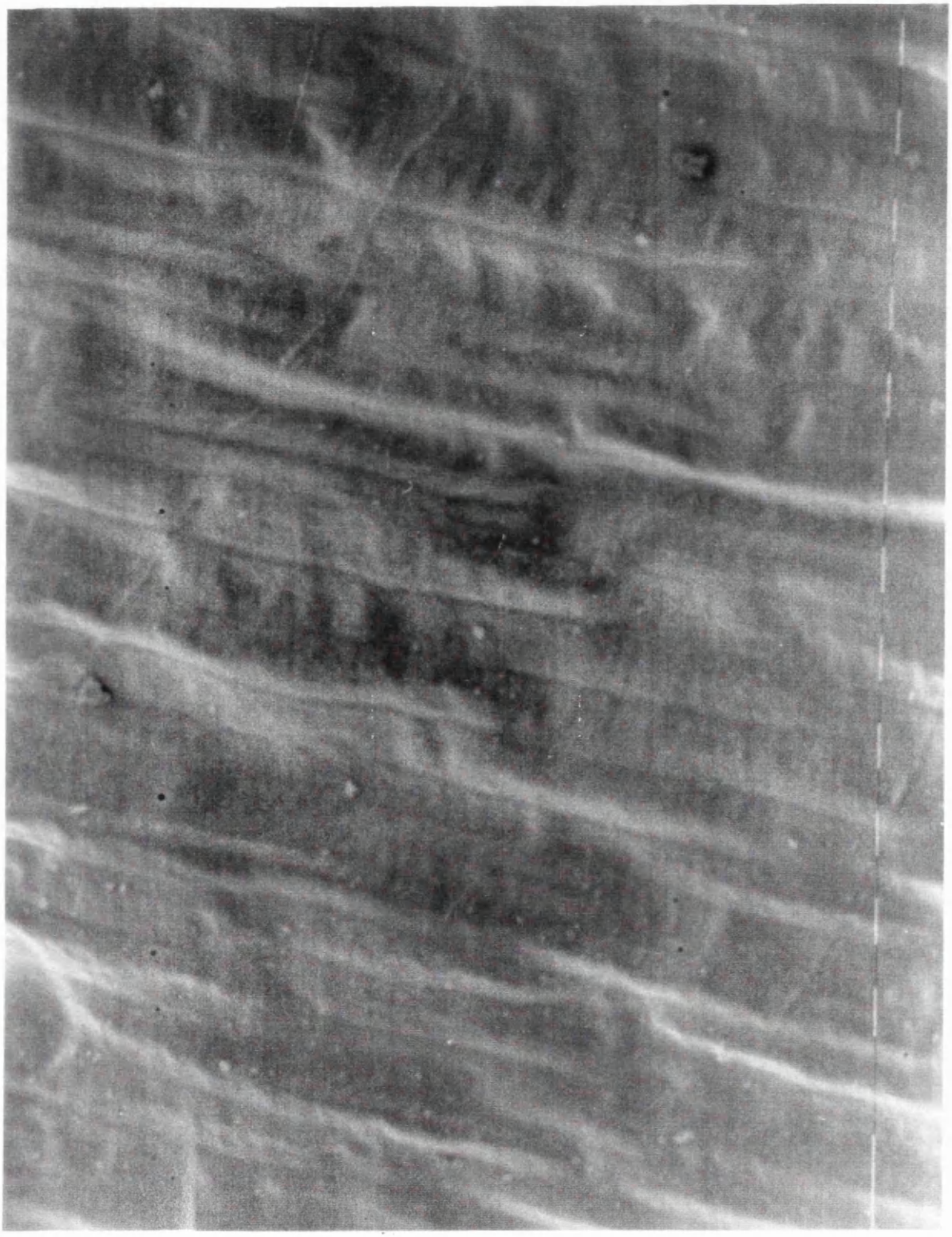


Figure 3.6 Scanning Electron Micrograph of Littorella surface scoring 5 on the Braun-Blanquet Scale. Scale bar: 1 division = 10 μm.

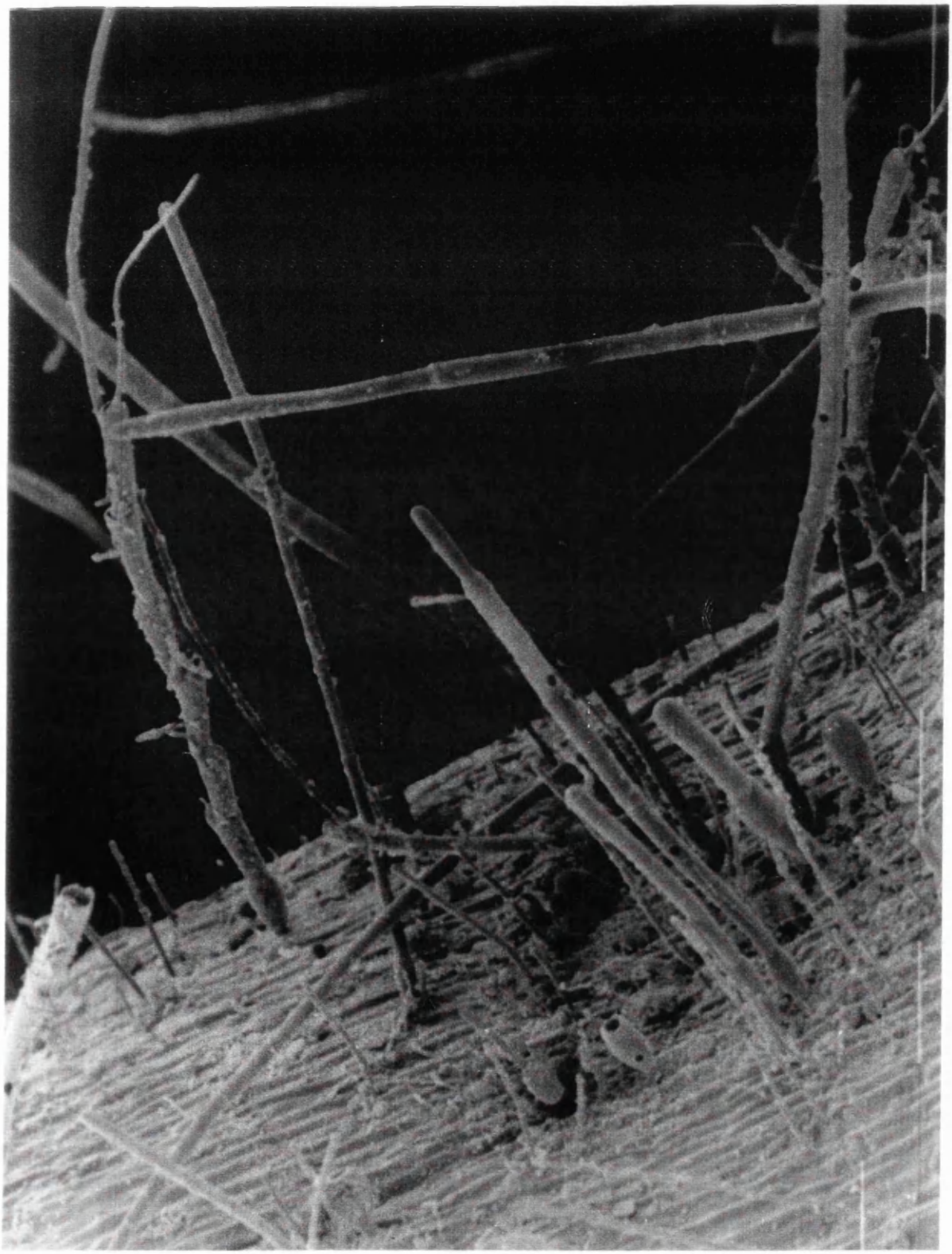
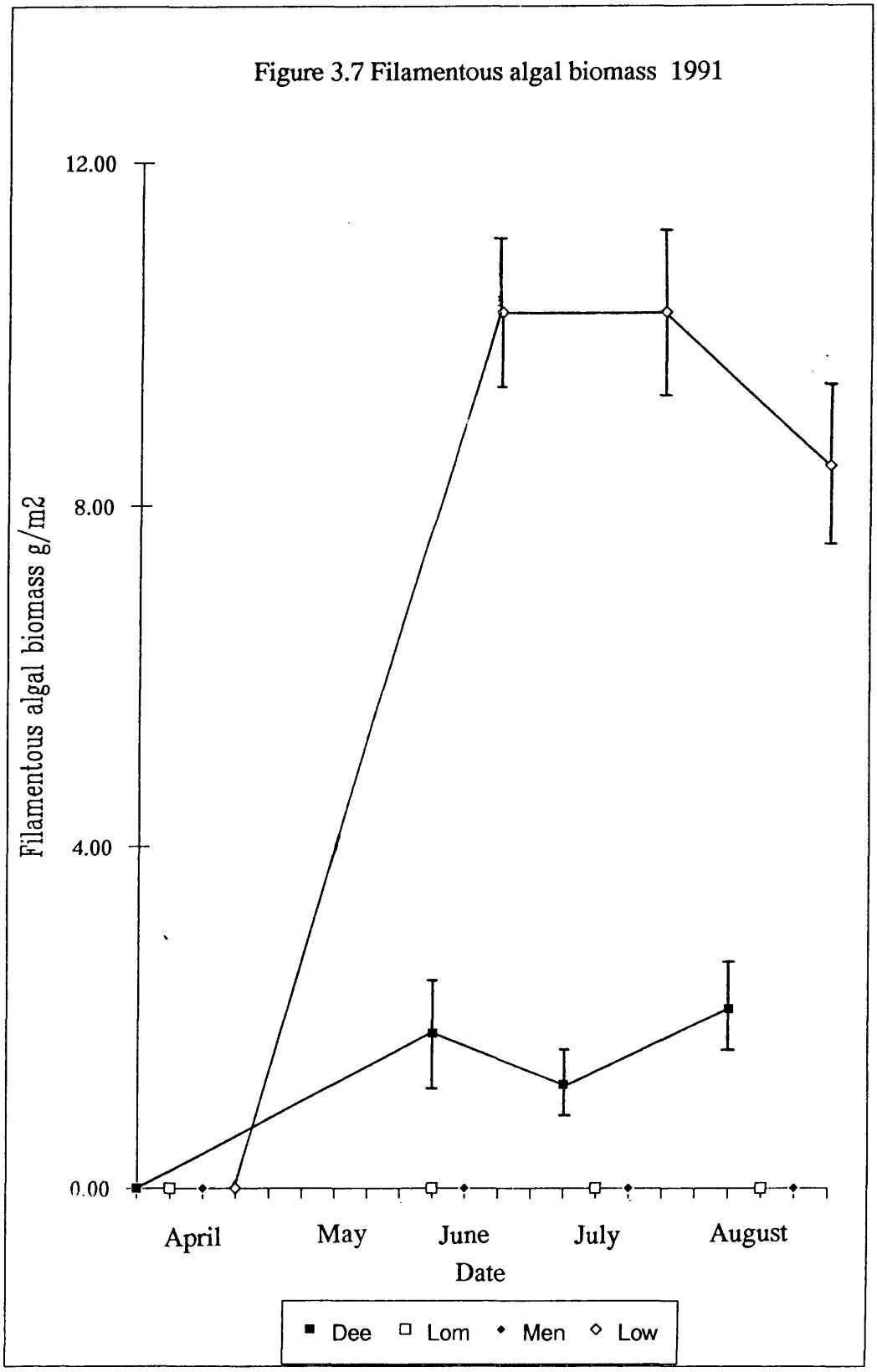


Figure 3.7 Filamentous algal biomass 1991



3.4 Filamentous Algae

Filamentous algal biomass was negligible at both Loch Lomond and the Lake of Menteith throughout 1990 and 1991. In Loch Dee and Loch of Lowes filamentous algal mats began to develop during May and were still evident by October during the 1990 sampling season.

In Lowes during June 1991 filamentous algal biomass reached a peak of about 10.3gm^{-2} with levels remaining high until September when sampling ceased. Filamentous algal biomass followed a similar pattern in Loch Dee with levels reaching a maximum of 2gm^{-2} - see Figure 3.7. Figure 3.9 shows the early stages of filamentous algal mat formation in Loch of Lowes.

The filamentous algal biomass in Loch Dee was dominated by *Oscillotaria* species, whereas those in both Lowes and Menteith were dominated by *Rhizoclonium* species. Due to the paucity of filamentous algae in Loch Lomond no samples were collected.

Section 4
DISCUSSION

The most commonly occurring species in all four lochs - *L. uniflora*; *I. lacustris*; *M. alterniflorum* and *J. bulbosus* - have all been described as ubiquitous in their distribution (Spence, 1967; Seddon, 1972).

E. canadensis was common in Loch Lomond samples in 1990, but less so in 1991 where the sampled areas were almost 100% *L. uniflora*. This species will rapidly colonise open sediments (Simpson, 1984) and was found in large patches in areas of exposed sediment caused by damage from boats.

P. praelongus was frequently found in the Lowes flora during 1990, but not 1991; this species is associated with a high siltation rate (Pearsall, 1918), reflecting the high siltation rates described in Loch of Lowes in Chapter 2, and is usually found in moderately rich lochs (Spence, 1967).

Trifonova (1989) proposed a scale for the classification of lakes in terms of their trophic state based on chlorophyll *a* concentrations. In this scheme the sites at Lochs Dee and Lomond would be oligotrophic, Menteith, with maximal chlorophyll *a* levels of around 10 µg l^{-1}, would be on borderline mesotrophic-eutrophic, and Loch of Lowes with chlorophyll a levels regularly in the region of 10 µg l^{-1} and a maximum recorded value of 16.6 µg l^{-1} in the autumn would be classified as eutrophic.

The incidence of filamentous cyanobacteria and green algal mat formation in Loch Dee and Loch of Lowes are typical of acidified and eutrophic lakes respectively (Hendry & Vertucci, 1980; Lazarek, 1985; Philips *et al.*, 1978).

Epiphyte colonisation on *Littorella* leaves was most dense on Lake of Menteith and Loch of Lowes samples. Although no measurements were taken of epiphytes on other species, it was apparent that colonisation on *Littorella* was less than on other plants such as *Elodea canadensis* and *Potamogeton* species; on no occasion was *Littorella* 100% covered in epiphytes. Moeller (1978) found isoetids did not, in general, support dense epiphytic growth.

Section 5
CONCLUSIONS

The four lochs represent variations of one community type, with some macrophyte species occurring in varying amounts in each of the four lochs e.g. *I. lacustris*; *M. alterniflorum*; *L. dortmanna* and *L. uniflora*. Environmental conditions range from acidic Loch Dee, to the oligotrophic mid basin of Loch Lomond, through to mesotrophic/eutrophic Lake of Menteith and eutrophic Loch of Lowes. As such the lochs exhibited a range of sediment types, water chemistry and algal loading.

Figure 3.8
Site 1 in Loch Lomond. Note the lack of filamentous algae, or phytoplankton.
Photographed by Alan Bell, June 1991.
Scale: Black line = 10cm.

Figure 3.9
Site 2 in Loch of Lowes, showing the early stages of filamentous algal mat formation. Photographed by Alan Bell, June 1991.
Scale: Black line = 10cm.

Chapter 4:

Attributes of *Littorella* Field Measurements

Chapter 4:

Attributes of *Littorella* Field Measurements

Section 1
INTRODUCTION

In Chapters 2 and 3 differences among the four study lochs were described, both in terms of physical, abiotic characteristics and their respective floras. Chapter 3 concluded that the four lochs studied represent a group of similar community types that ranged from acidic Loch Dee to moderately eutrophic Loch of Lowes. The following chapter presents data on the morphology and some chemistry of *Littorella* in the four lochs and relates this to the measured abiotic factors and algal loading of the four lochs described in Chapters 2 and 3.

Section 2
METHODS

The following measurements were carried out on *Littorella* plants collected in the sampling regime described in Chapter 3: total biomass: percentage abundance: number of leaves: leaf length: weight of 5 plants (1991): leaf area ratio (1991): root:shoot: stolon length: number of plants per m^2 (1991): number of stolons per plant (1991): nitrogen and phosphorous content (1991): total chlorophyll concentration: chlorophyll *a:b* (1991). Unless otherwise stated, data were obtained for both the 1990 & 1991 growing seasons.

Throughout this chapter the term plant has been used to describe an individual *Littorella* rosette. During 1990 photosynthetic measurements on *Littorella* plants collected from the field were attempted. Due to practical difficulties (technical difficulties with equipment resulted in problems getting reproducible results) this was not carried out in 1991 . The protocol used and results obtained are presented in Appendix D5 for interest.

2.1 Total Biomass and % Abundance

Biomass samples were collected with the aid of SCUBA and dried in an oven at 90°C, as described in Chapter 3. Percentage abundance of *Littorella* was expressed on a dry weight basis.

2.2 Number of leaves, leaf length, surface area and weight of five plants

Five plants were selected from each quadrat, from each plot, on each site visit during 1990 & 1991 (n=160 per plot; total n=1920). The number of leaves on each plant were counted, their length measured in mm, and the diameter was measured at three points to 0.1mm tolerance using a stage graticule and dissecting microscope. The three measurements of diameter were carried out at three different points in the same location along the length of each leaf measured. The leaf surface area was estimating by assuming the leaf to be a tube and using the following equation:

$$\text{Area} = \pi.D.L$$

Where π = 3.1416
D = the mean diameter
L = length of leaf.

After these measurements were carried out the five plants were dried at 90°C and weighed.

2.3 Leaf Area Ratio

Leaf area ratio (LAR) was calculated by dividing the total leaf area of five plants by their weight. LAR is a morphological measurement of the leafiness of a plant, as the potential photosynthetic tissue is compared with the potential respiring parts of the plant (Hunt, 1990).

2.4 Root:Shoot Ratio (R:S)

A sample of total biomass was selected (between 50 and 100% of the total sample depending on sample size), and then divided into root and shoot tissue, dried as described in Chapter 3 and weighed.

2.5 Number of *Littorella* plants per m^2

This figure was estimated by multiplying the total *Littorella* biomass for $0.0625m^2$ by 16. The resultant biomass per m^2 was divided by the weight of five plants and then multiplied by 5 to get the number of plants per m^2.

2.6 Stolon length and number of stolons per plant

During 1990 the length (mm) of all stolons observed was measured. During 1991 the entire sample was sorted, the number of stolons were counted and the length of the first 50 observed were measured. Broken stolons were counted as long as the break had occurred at one end of the stolon and the longest part had been recovered. The location of the break is obvious from the fact that the stolon is broader at either end than in the middle.

Number of stolons per plant was estimated by multiplying the total number of stolons per $0.0625m^2$ by 16 and the dividing this figure by the estimated number of plants per m^2.

2.7 Nitrogen and phosphorous concentration

The Kjeldahl method allows quantitative analysis of nitrogen and phosphorus from plant material in one digest. This method generally involves a digestion phase, to convert organic compounds to inorganic compounds, followed by a determination of the concentration of inorganic ions in the digest (Nelson & Summers, 1973).

The digestion procedure consists of heating a small amount of sample with a concentrated strong acid (e.g. sulphuric acid) with salt (e.g. potassium sulphate

added to raise the boiling temperature), in the presence of a heavy metal catalyst such as selenium, copper or mercury.

In this case a copper catalyst was selected although it has a lower yield than selenium (Nelson & Summers, 1973), as selenium was found to form black deposits on the top of the Kjeldahl tubes. Mercury was not used as it is a hazardous substance and copper is less toxic.

To increase the recovery of nitrate-N and nitrite-N a pre-treatment which involved warming the dried plant sample with salicylic acid and sodium thiosulphate was carried out (Bremner & Mulvaney, 1982).

Samples were digested in a block digestor, which allows the simultaneous digestion of a large number of samples and good temperature control of the digestion stage (Bremner & Breitenbeck, 1983).

2.7a **Sampling and pre-treatment**

Fresh leaf and root material from entire *Littorella* plants collected during the final sampling visits of 1991 was rinsed in distilled water then dried in an oven separately at 60°C. After drying, the leaf and root material were ground in a coffee grinder (Braun Aromatic KSM2) until the material was fine enough to pass though a 40 mesh sieve (Nelson & Summers, 1973).

Plant material was collected in August/September 1991. Equal quantities of dried material from each plot were pooled to make one sample per site. This material was then stored in an airtight container in darkness until analysis could be carried out (after about one month).

2.7b **Pre-digestion**

A sub-sample of ground plant material was accurately weighed (± 0.0001g) out into a small pre-weighed aluminium foil vessel. This was lowered into the bottom of a Kjeldahl boiling tube and emptied, making sure that none of the plant material stuck to the sides of the tube. The aluminium vessel was then re-weighed to correct for any material that may not have been dispensed into the boiling tube.

5ml of salicylic acid dissolved in concentrated hydrochloric acid (250g l^{-1}) was added to the plant material and allowed to stand for one hour. Five hundred milligrams of sodium thiosulphate pentahydrate was then added to the boiling tubes and the tubes were warmed gently, shaken and left to cool. One gram of sodium sulphate and copper (II) sulphate mix (10:1 by weight) was then added.

2.7c **Digestion**

The tubes were lowered into a preheated (375°C) block digestor in a fume cupboard. A rack of tube tops connected to a condenser supplied with tap water was placed on top of the boiling tubes, and the tubes were left to boil for 180 minutes. The tubes were then removed from the digestor with tops intact and allowed to cool to room temperature. The solution was diluted to 100ml with deionised water and shaken thoroughly to dissolve the contents. Digests were then filtered and a middle 20ml fraction was collected and stored in a clean (washed in dilute hydrochloric acid) screw topped polypropylene container.

Three control measures were carried out during the digestion procedure.

1. Two digests were carried out on each sample.
2. A blank was digested where no plant material was added.
3. 1ml of 1% (w/w) ammonium-N was added after the pre-digestion stage to a sample containing no plant material.

No nitrogen was detected in the blank sample and constant values of ± 0.05% were found within the replicated samples for the ammonium-N control.

Ammonium-N and phosphorus concentration in the digests were determined using a Technicon Autoanalyser II colorimeter. Results are expressed as a percentage of the dry weight of *Littorella.*

2.8 Leaf Chlorophyll concentration

Chlorophyll concentration was measured by extraction in hot methanol using an adaptation of the method described in Hipkins & Baker (1986). Extraction was tested with hot and cold methanol and hot and cold acetone. Hot methanol (70°C) was found to extract the chlorophyll most rapidly. The rationale behind the use of methanol as an extraction solvent are discussed in Chapter 3, Section 2.3.

The following protocol was carried out in dim light in order to minimise photodegradation of chlorophyll.

Tissue was selected from healthy plants, the two youngest leaves were selected if the smallest leaf was greater in length than 1cm. Tissue of similar age was used in order to allow comparisons to be made between samples (Farmer & Adams, 1989). Plant material was cleaned of any epiphytic growth by washing under the tap and whilst rubbing gently between forefinger and thumb. Microscopic examination using a Nikon Binocular dissecting microscope revealed there to be few epiphytes remaining after such washing. After washing, the leaves were blotted dry to remove any residual water.

The two leaves were then cut vertically using a clean, new razor blade, and cut into 2mm sections. Cutting the leaves greatly improved the recovery of chlorophyll with some pigment still apparent in the tissue of intact leaves after 6 hours of extraction.

The cut tissue was then added to 5-to-10ml of analytical grade methanol in a foil wrapped centrifuge tube then placed in a water bath at 65°C for 10 minutes.

The methanol was removed and a further aliquot (less than 5ml) of methanol was added and the extraction was repeated. The resultant volume from this extraction

was added to the first extract volume. After two such extractions the tissue appeared completely colourless. The chlorophyll/methanol solution was allowed to cool to room temperature and then made up a to a known volume in a volumetric flask. This was then centrifuged at 3,000 g for five minutes in order to remove any particulate matter.

Absorbance was measured at 665 and 650nm using a Shimadzu UV-160A UV-visible recording dual beam spectrophotometer (bandwidth 2nm). Chlorophyll concentrations were calculated using the following equations:

$$\text{Chl a} = 16.5A665 - 8.3A650$$
$$\text{Chl b} = 33.8A650 - 12.5A665$$
$$\text{Total Chl} = 25.8A650 + 4.0A665$$

2.9 **Data Analysis**

In order to determine if there was any relationship between the attributes measured in this chapter and the environmental parameters described in Chapters 2 & 3, stepwise multiple linear regression was carried out using the statistics package SPSS. This technique allows the changes in an attribute to be described in terms of independent variables. The variability of each observation is due to the sum of all sources of variance. Therefore, if all sources of variance, such as the important environmental factors, are accounted for, the residual variation should be zero (Sokal & Rohlf, 1981).

Data collected over the two years were analysed separately. In the 1990 data set, only data obtained in the last three field visits were used due to the change of sampling technique employed after the first visit. The mean value of each attribute for each site visit was included in the analysis (a total of 47 values in 1991 - Loch Dee site 2 first visit data omitted due to bad weather during sampling - and 36 in 1990).

Due to large daily variations in temperature and percentage oxygen saturation these two variables were not included in the data analysis. In order to consider seasonal

effects, sampling date was included as an independent variable. Missing variables were replaced by the mean of the remaining measured values for the same site. No more than one variable per site was omitted, and 80% of the measured variables were complete. Independent and dependant variables used in the 1990 and 1991 data analysis are listed in Table 4.1. Equations obtained from the regressions are presented in Tables 4.2 and 4.3 for 1990 and 1991 respectively.

As nitrogen and phosphorus concentration in the roots and shoots were measured on the last sampling visit only these data were analysed separately and regressed against environmental data collected on the same dates as the plant samples were collected.

Table 4.1a
Independent variables used in multiple regression

variable	year	abbreviation
pH	90/91	pH
conductivity	90/91	COND
phytoplankton chlorophyll *a*	90/91	CHLa
filamentous algal biomass	91	FILALG
epiphyte percentage cover	91	EPIP
exposure	90/91	EXP
extinction coefficient	90/91	E
sedimentation rate	90/91	SEDR
sediment organic content	90	SEDORG
sampling period	90/91	DATE

Table 4.1b
Dependent variables used in multiple regression

variable	year	abbreviation
total leaf chlorophyll	90/91	LFCHL
chlorophyll *a:b*	91	CHLa:b
stolon length	90/91	STL
no. stolons per plant	91	STNO
leaf number	90/91	LFNO
leaf length	90/91	LFL
root shoot ratio	90/91	R:S
percent abundance	90/91	PA
no. plants per m^2	91	LITTNO
weight 5 plants	91	WT5
leaf area ratio	91	LAR
total biomass	90/91	TOTBIO
Littorella biomass	90/91	LITTBIO
% leaf nitrogen	91*	LFN
% leaf phosphorous	91*	LFP
% root nitrogen	91*	RN
% root phosphorous	91*	RP

*Last sampling visit in 1991 only

Section 3
RESULTS

Littorella attribute data used in the multiple stepwise linear regression are tabled in Appendix D3. Sampling date was selected as a predictive variable for two *Littorella* attributes. In 1990, 13.8% of the variation in root:shoot was explained by the sampling period. In the 1991 samples no variable was selected to explain the variation in this data. In 1991, sampling date explained 14.2% of the variation in stolon length data, suggesting that as the growing season progressed stolons became longer. However in 1990, 16.1% of the variation of this attribute was related to phytoplankton chlorophyll *a* levels. The fact that sampling date was not selected as a predictive variable suggests that seasonal variation was outweighed by the effect of other measured environmental variables.

In 1990, 18.1% of the variation in *Littorella* total leaf chlorophyll content was explained by the presence of phytoplankton chlorophyll *a*. However, in 1991 filamentous algal biomass explained 60.2% of the *Littorella* total leaf chlorophyll, with an increase in filamentous algae resulting in higher leaf chlorophyll content, the influence of chlorophyll *a* explained a further 5.1%. The 10.7% variation in chlorophyll *a*:*b* was also explained by filamentous algal biomass, with chlorophyll *b* increasing as filamentous algal biomass increased.

In 1991 filamentous algal biomass explained 18.7% of the variation of number of stolons per plant and 21.6% of the variation in leaf length, with more stolons and fewer leaves being formed in the presence of filamentous algae.

Leaf number in 1990 was related to extinction coefficient, as was leaf length (percentage variation explained = 12.7 and 24.7 respectively), whereas the most important measured variable to explain variation in leaf length in 1991 was water conductivity.

Percentage abundance was related to exposure rating in 1991, and to chlorophyll *a* in 1990. In the 1991, sites with a high exposure rating tended to have fewer

smaller plants. Exposure rating explained 12.0% and 25% of the variation in the number of plants per m^2 and weight of 5 plants respectively.

Variation in macrophyte total biomass and *Littorella* biomass were accounted for by the sediment organic content in 1990. When soil organic content was removed, there was no predictive variable for total biomass, whereas 17.5% of the variation in the *Littorella* biomass could be explained by the presence of phytoplankton as estimated by chlorophyll *a*. In the 1991 data set 27.1% of the variation in the data could be predicted by the exposure rating of the site, no independent variables were selected to predict *Littorella* biomass. From these results it would appear that the most important factors influencing submerged aquatic macrophyte biomass in the four lochs studied were exposure and sediment organic content.

Forty-eight percent of the variation in leaf nitrogen was explained by filamentous algae. The residual variation in leaf nitrogen content was best explained by the extinction coefficient, which accounted for a further 26.9% of the variation. No independent variables were selected to explain either the variation in root nitrogen, or root and leaf phosphorous.

A summary of the primary environmental variables selected by the stepwise multiple linear regression is presented in Table 4.4 at the end of this chapter.

Table 4.2

Equations obtained from stepwise linear multiple regression of 1990 data

Equation	% variation explained
LFCHL = 15.0 + 0.271 CHLa	18.1**
STL = 88.8 - 1.06 CHLa	16.1*
LFNO = 4.31 + 0.27 E	12.7*
LFNO = 1.73 + 0.32 E + 0.385 pH	24.7**
LFL = 40.8 - 0.303 EXP	13.7*
R:S = 0.881+0.0925 DATE	13.8*
PA = 74.3 - 2.63 CHLa	20.7*
TOTBIO = 59.0 + 7.95 SEDORG	11.0*
LITTBIO = 21.5 + 8.27 SEDORG	36.1***

Unselected independent variables:

1990	1991
COND; SEDR	EPIP

* $P < 0.05$

** $P < 0.01$

*** $P < 0.001$

Abbreviations as listed in Table 4.1b

Table 4.3

Equations obtained from stepwise linear multiple regression of 1991 data

Equation	% variation explained
LFCHL = 13.13 + 0.912 FILALG	60.2****
LFCHL = 12.40 + 0.741 FILALG + 0.399 CHLa	65.3****
CHLa:b= 3.13 - 0.475 FILALG	10.7*
STL = 58.2 + 7.56 DATE	14.2**
STNO = 0.214 + 0.023 FILALG	18.7**
LFNO = 5.25 - 0.843 FILALG	21.6**
LFNO = 3.62 - 0.108 FILALG + 0.249 pH	30.9***
LFL = 49.36 - 0.235 COND	26.3***
R:S = no independent variable selected	
PA = 83.2 - 1.01 EXP	18.0**
PA = 77.1 - 1.25EXP + 26.5 SED	25.1**
LITTN = 2284 - 37.21 EXP	12.0*
LITTN =	
WT5 = 0.186 - 0.0029 EXP	12.0*
WT5 = 0.193 -0.0027 EXP - 0.0049 FILALG	32.2***
LAR = 3.33 - 0.634 SEDR	9.1*
TOTBIO = 9.46 - 0.206 EXP	27.1***
LITTBIO = no ind. variable selected	

Abbreviations as listed in Table 4.1b

Table 4.3b
Equations obtained from stepwise linear multiple regression of 1991 final sample visit only

Equation	% variation explained
LFN = 2.00 + 0.O457 FILALG	48.1*
LFN = 1.15 + 0.0615 FILALG - 0.983 E	75.0**

LFP = no independent variable selected

RN = no independent variable selected

RP = no independent variable selected

* $P < 0.05$
** $P < 0.01$
*** $P < 0.001$
**** $P < 0.0001$

Abbreviations as listed in Table 4.1b

Section 4
DISCUSSION

The ecological relationships identified from the field data cannot be used to infer causal relationships between *Littorella* attributes and environmental parameters. Such regression relationships can be used, however, to generate hypotheses concerning the ecological interactions of *Littorella* attributes with environmental factors. The hypotheses discussed in the following section were generated from relationships identified in the field data. Some hypotheses will be tested in subsequent chapters.

In general, the percentage variation of morphological attributes of *Littorella* accounted for the measured environmental parameters is low. Wilson (1991) measured morphological traits of 12 lake shore plants along an environmental gradient of sediment organic content. Variations in morphology in an experimental gradient of sediment nutrient concentration were measured and compared to the plasticity of field measurements of the 12 plants. *Littorella* was not included in the study, however in rosette-forming plants, such as *Lobelia dortmanna,* the maximum variation of morphological attributes accounted for by regression against organic content were 23% and 22% for biomass and R:S respectively. In the majority of other morphological traits only 10-15% of the variation could be accounted for. Non-rosette species showed the greatest plasticity in morphology, with up to 80% of variation in morphological attributes being explained by the position along the experimental gradient of soil organic matter. Wilson (1991) concluded that rosette-forming species adjust physiologically, rather than morphologically, to changes in the environment.

In the work presented here, a far greater variation in physiological attributes, such as total chlorophyll content and shoot nitrogen content (65% and 75% respectively), were accounted for by the measured environmental parameters. Grime *et al.* (1986) stated that stress tolerant plants which may be characterised by evergreen species that have slow relative growth rates and which depend primarily on vegetative means of reproduction, will give rise to small changes in morphology with changing environmental conditions. Such 'stress-tolerating' species will tend

to adapt to changes in the environment by physiological means. Holstrup & Wiegleb (1991b) observed that over a six month period, there were no changes in the morphology of *Littorella* under conditions of low nutrients or low irradiance.

Macrophyte biomass per unit area has been described as a relative measure of habitat colonisation and relative species importance within a lake (Moeller, 1975). In 1990, both total biomass and *Littorella* biomass increased with increasing sediment organic content. Szmeja (1987a) reported low macrophyte biomass in areas of low sediment organic content in isoetid-dominated lakes in Poland. In 1991, in the absence of sediment organic measurements, there was no environmental parameter selected to account for the variation in the *Littorella* biomass, and total macrophyte biomass decreased with increased exposure to wind/wave action. Of the environmental parameters measured in the four lochs studied, the most important determinant of *Littorella* biomass was organic matter concentration.

In the 1991 data set, the measured environmental parameters most frequently selected by the stepwise multiple regression in the four lochs studied were exposure and filamentous algal biomass. Collins *et al.* (1987) measured the distribution of 38 aquatic macrophyte species in relation to 4 depth classes and 13 physical parameters. By the use of factor analysis and ordination, these workers found depth, substrate type and eutrophication to be primary correlative factors in macrophyte distribution. As all the measurements in this study were carried out along the 1m isobath, depth is not relevant. Substrate type is often related to exposure (Keddy, 1982) and eutrophication has been related to algal biomass (e.g. Phillips *et al.*, 1978).

Increased exposure to wind/wave action, as estimated by exposure index (Weisner, 1987), resulted in fewer, smaller *Littorella* plants with a reduced percentage abundance of a smaller total macrophyte biomass. In 1990, increased wind/wave exposure resulted in *Littorella* plants with shorter leaves, although in the 1991 data set when filamentous algal biomass was measured, this reduction in leaf length was related to the presence of filamentous algae.

Farmer and Spence (1987) found *Lobelia* plants growing in more exposed sites to be of smaller size, than those in sheltered sites. However, the difference in sediment characteristics between exposed and sheltered sites makes it difficult to distinguish between mechanical effects due to exposure, and effects due to sediment characteristics. In this study the most sheltered sites (i.e. those in Loch Lomond) were also the sites with the highest organic sediment content (see Table 2.2). Gacia and Ballesteros (1993) attributed the smaller size of individuals of *Isoetes lacustris* sampled from shallow water, when compared with deep water individuals, to a fast turn-over rate. These workers attributed a fast turn-over of individuals in shallow water populations to disturbance due to wave action and ice formation rather than the light regime, although sediment organic content effects could not be ignored. The idea of a faster turn-over of individuals in shallow water is supported by the work of Szmeja (1987c), where deep water populations of *L. dortmanna* were observed to consist of more mature individuals and fewer juveniles than shallow water populations.

The presence of filamentous algae accounted for an increase in leaf chlorophyll and nitrogen, and a decrease in chlorophyll *a:b*. Such changes in chlorophyll concentration are typical of a shade response (Björkman, 1981).

In the absence of filamentous algal biomass measurements from the 1990 data set, phytoplankton chlorophyll *a* levels accounted for variation in the *Littorella* leaf chlorophyll. In 1991, phytoplankton chlorophyll *a* was not selected as a primary source of variation in any of the measured *Littorella* field attributes. In 1990, phytoplankton chlorophyll *a* was the main environmental variable selected to account for the variation in 3 out of 8 measured *Littorella* field attributes. From these data it would appear that, in the absence of filamentous algal biomass data, the influence of phytoplankton may be over-emphasised.

Søndergaard and Bonde (1988) calculated that epiphyte shading on *Littorella* reduced irradiance by 10-to-24%. Robe and Griffiths (1992) reported up 90% shading by filamentous algal mats. Figure 3.6 (p58) shows a scanning electron micrograph of one the areas most densely colonised by epiphytes. Sand-Jensen (1977) in a study on eel grass, demonstrated that as well as reducing irradiance,

epiphytic diatoms acted as a barrier to inorganic carbon diffusion. In species such as *Littorella*, where carbon uptake is primarily through the roots, epiphyte effects on diffusion are of less importance. In comparison to other algal groups, epiphytes had little effect on *Littorella* in the four lochs studied.

Søndergaard and Bonde (1988) found there to be no seasonal variation in the total chlorophyll, chlorophyll *a*, chlorophyll *b* concentrations and chlorophyll *a:b* in leaves of *Littorella* growing at 0.2m depth in an oligotrophic lake until November when the incident irradiance decreased. In the same study deep water (2.3m) plants only increased in total chlorophyll and chlorophyll *a* in November. Deep water plants had significantly higher levels of total chlorophyll, chlorophyll *a*, chlorophyll *b* and a higher chlorophyll *a:b* than shallow water plants throughout the study period. Sand-Jensen (1978) found there to be no significant seasonal change in *Littorella* chlorophyll content throughout the year, although he did note a trend to higher concentrations in the winter. Both these studies were carried out in lochs without high algal loading.

Lazarek (1986) in a study on *L. dortmanna* found there was an increase in leaf chlorophyll after the loss of epiphytes, which occurred three years after liming of an acidified loch. However, post-liming measurements were carried out in August and pre-liming measurements in July. The incident PAR levels were considerably lower in the period prior to the post-liming measurements, so changes in chlorophyll concentration could not be attributed to epiphyte effects.

The nitrogen and phosphorus concentrations measured in the leaves of *Littorella* are comparable to values obtained by other workers (Moeller, 1978; Allenby, 1981). Root nitrogen content was higher than the figures for *L. dortmanna* reported by Moeller (1978), but was in agreement with those reported for *Littorella* by Robe & Griffiths (1992). Robe & Griffiths, from their comparison of *Littorella* plants from an oligotrophic and eutrophic site, found the nitrogen content in leaves of *Littorella* to be higher in plants sampled from the eutrophic site. They attributed this difference to the greater chlorophyll content and higher photosynthetic capacity of these plants. A similar response in terms of total chlorophyll content was observed in *Potamogeton pectinatus* by Hootsmans & Vermaat (1991). These

authors, however, noted that there were practically no differences in the chlorophyll *b* fraction and nitrogen content of plants grown at high and low irradiance.

In the presence of filamentous algal biomass, *Littorella* plants had a greater number of stolons per plant and fewer leaves. The increased allocation to reproduction is contrary to what Grime *et al.* (1986) predicted for small evergreen plants such as *Littorella*, where under conditions of stress the main energy investment would be in maintenance of the adult plant rather than to reproduction. From work in the Lake District (England), Robe & Griffiths (1992) reported that *Littorella* plants in eutrophic Esthwaithe Water had more ramets per plant and more short leaves compared with plants sampled from acidic Red Tarn. Both these sites experienced filamentous algal mat formation and data from transplant experiments suggested that differences in ramet production were due to genetic differences in the two populations.

Farmer & Spence (1986) suggested that the success of *Littorella* in more eutrophic waters, in comparison to other isoetids, may be due to their faster growth rate and stolon production, which may enable *Littorella* to outgrow epiphytes. However, Nielsen & Sand-Jensen (1991) suggested that *Littorella* was excluded from eutrophic lakes due to shading by algae and the competitive effects of more vigorously growing rooted macrophytes.

As *Littorella* obtains the bulk of its inorganic carbon from the sediment (Sand-Jensen & Søndergaard, 1978), the main effect of the presence of algae will be due to shading. The next two chapters consider the effects of shading and sediment organic content on the morphology and physiology of *Littorella* under controlled conditions. Chapter 5 considers the effects of sediment organic content and shading on the morphology and chlorophyll content of *Littorella*. Chapter 6 considers the physiological adaptation of *Littorella* to shade.

Table 4.4
Summary of primary environmental variables selected by stepwise multiple linear regression

	COND	CHLa	FILALG	EXP	E	SEDR	SEDORG	DATE
LFCHL		-	+					
CHLa:b			-					
STL		-						+
STN			+					
LFNO			-		+			
LFL	-				+			
R:S								+
PA		-		-				
LITTN				-				
WT5				-				
LAR						-		
TOTBIO				-			+	
LITTBIO							+	
LFN			+					
TOTALS	0 1	3 0	* 5	1 4	2 0	0 1	2 *	1 1

Key:
1990 data on box left, 1991 data on right
- negative influence
+ positive influence
Totals are the total number of times the environmental variable has been selected as accounting for the greatest variation in the measured attributes.
* parameter not measured in that year
Abbreviations as listed in Table 4.1a

Section 5
CONCLUSION

In the absence of filamentous algal biomass measurements during the 1990 season, phytoplankton chlorophyll *a* was the most commonly selected environmental variable that accounted for variation in the 1990 data set. *Littorella* plants growing in the presence of higher chlorophyll *a* levels had higher leaf chlorophyll, shorter stolons and a lower percent abundance. Both *Littorella* and total macrophyte biomass were positively related to sediment organic content.

In the 1991 data set, the two most important variables in determining *Littorella* attributes were exposure rating and filamentous algal biomass. *Littorella* plants growing under filamentous algal mats tended to have a higher total chlorophyll content a greater number of stolons per plant, a higher leaf nitrogen content, fewer leaves and a lower chlorophyll *a*:*b*. Epiphytes did not have a significant effect on the measured attributes of *Littorella* in the four lochs studied.

More exposed sites tended to have fewer *Littorella* plants of smaller size (dry weight) and had a lower percentage abundance and total biomass than more sheltered sites.

Changes in the chlorophyll content of leaves of *Littorella* indicate a possible shade response. The following Chapters report experiments on the shade response of *Littorella* in controlled greenhouse conditions in order to determine whether similar attribute changes to those observed in the field also occur in plants subjected to shading alone.

Chapter 5:

Effects of Shading on the Growth of *Littorella* - Greenhouse Trials

Chapter 5:

Effects of Shading on the Growth of *Littorella* - Greenhouse Trials

Section 1
INTRODUCTION

Field observations presented in Chapter 4 demonstrated morphological differences in *Littorella* plants collected from the same water depth at different sites. Size differences in *Littorella* were primarily attributed to exposure rating and sediment organic content. Results from stepwise linear multiple regression of *Littorella* attributes against environmental data (Chapter 4) selected filamentous algal biomass to account for most of the variation in the number of leaves and number of stolons per plant. The work described in this chapter investigated the effects of shading and sediment organic content on the morphology of *Littorella.*

The first experiment tested the effects of different sediment organic matter concentrations on the growth of individual *Littorella* plants. Results from this experiment were used to determine the optimum sediment organic matter content for subsequent greenhouse experiments.

Filamentous algal mats were not present in Loch of Lowes and Loch Dee throughout the year. It is hypothesised that *Littorella* populations can be maintained throughout periods of low light intensity, and increase biomass under conditions of higher irradiance that exist before and after the formation the algal mat. The second experiment presented in this chapter, tests the effects of shading and subsequent removal of shading on the morphology and chlorophyll content of *Littorella.*

Section 2
METHODS

All plant material used in these studies was taken from greenhouse stock that had originally been collected near the study site in Loch Lomond in October 1989.

2.1 Macrophyte Culture System

All greenhouse experiments were carried out in a set of interconnected tanks through which tap water was recirculated. The water in each tank was aerated to encourage macrophyte growth and reduce epiphytic algae (Robson, 1974).

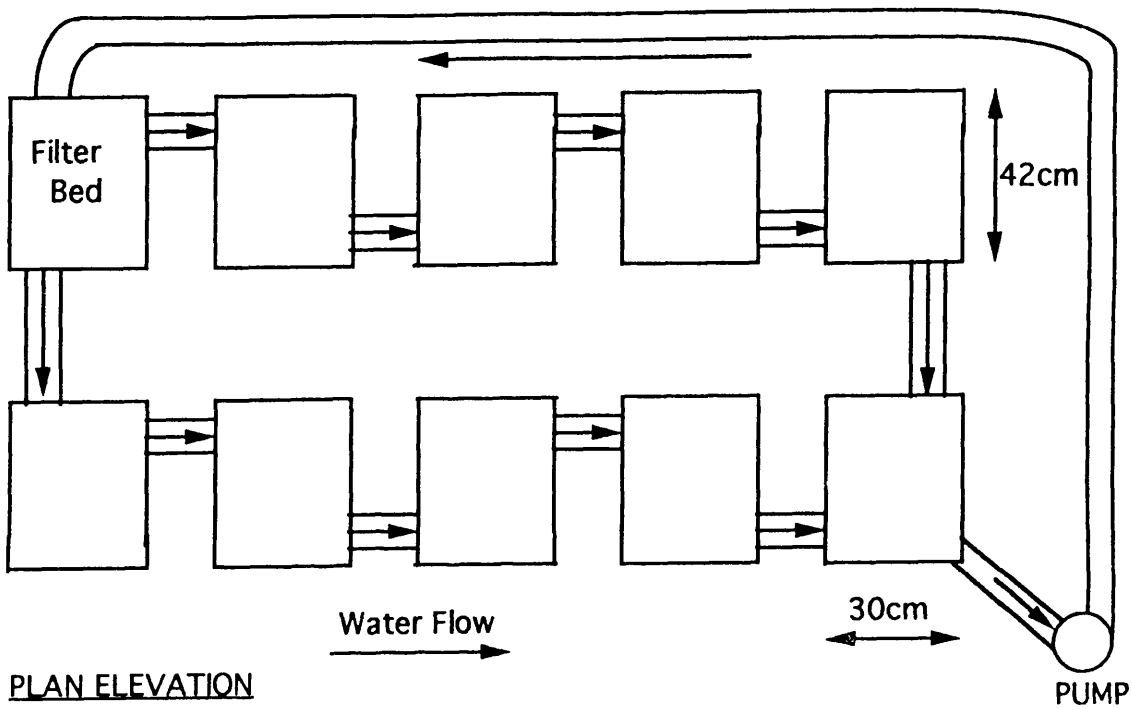
Ten, 40 litre water storage tanks were connected in a circular series by 2.5cm diameter plastic piping. Inlets and outlets were positioned alternately at the top and bottom of each tank to encourage mixing of the water column and prevent stratification (Figure 5.1).

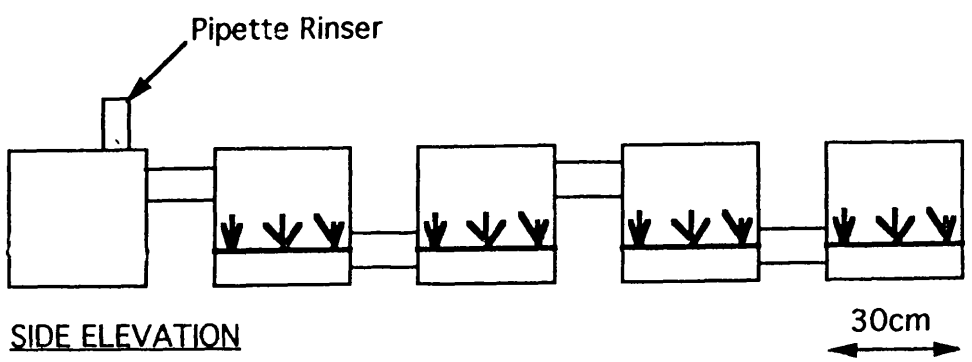
An electric pump (Eheim 1121) carried water from one end of the system to the other, where it was filtered through a gravel bed. The gravel bed consisted of a pipette cleaner (height 50cm; diameter 13cm) filled with stones of an approximate diameter of 1-2 cm, placed on top of a gravel-filled cold water tank. The pipette cleaner ensured that the water flow was in pulses, so preventing a head of water building up at one end of the tank series. In order to prevent algal growth the filtering system was shaded by black polythene.

The whole system had a volume of 400 litres and was recirculated approximately four times daily. Losses of water by evaporation were replaced by a slow drip of water at the pump end, which ensured that a constant water level was maintained. Individual tanks could be isolated for routine maintenance and cleaning by fitting 'suba-seal' rubber stoppers over the inlet and outlet of the tank to be isolated.

This set-up ensured minimal differences in water quality between treatments and was effective in reducing unwanted epiphytic and phytoplankton growth.

FIGURE 5.1 Schematic Diagram of Macrophyte Culture System.





2.2 The Effect of Sediment Organic Content on the growth of *Littorella*

Individual *Littorella* plants of similar size (leaf number = 4) were planted in 76mm diameter pots containing the following proportions of sand and peat:

% peat	% sand
0	100
10	90
25	75
50	50
25	75
100	0

The sand was collected from the oligotrophic north basin of Loch Lomond at Rowardennan and washed in tap water to remove organic matter. The peat used was moderately rotted sphagnum peat (ICI). Ratios were prepared on a volume:volume basis.

Four replicates from each sediment mix were set up and placed in one of four cold water storage tanks, so that each contained one of each of the four above mixtures. Pots were rearranged every two weeks to minimise possible shading effects due to the sides of the tanks. The experiment was run from October to March and natural daylight was supplemented with mercury lamps set to have an 18 hour photoperiod. Maximum unshaded light intensity was in the region of 473 $\mu E\ m^{-2}\ s^{-1}$ (measured using a Skye SKP200 PAR meter at mid-day on an overcast day in February).

After five months the plants were harvested and the following parameters measured using the methods described in Chapter 4: chlorophyll content: dry weight: root:shoot dry weight ratio: Leaf Area Ratio (LAR).

The Specific Leaf Area (SLA) was also measured on plants harvested in this chapter. SLA was calculated by dividing the total area of the plant shoot by the

total leaf dry weight biomass (Hunt, 1990). Specific leaf area has been shown to increase on shading in a number of aquatic macrophytes (e.g. Spence & Chrystal, 1970b).

2.3 Effects of Quantitative Shading on the Morphology and Leaf Chlorophyll Content of *Littorella*

Individual plants of *Littorella* were placed in 50:50 peat:sand as described in section 2.2. Six pots were placed in each of 6 greenhouse tanks and left for a period of two weeks to acclimatise. After this period, shading was applied to three of the tanks in he form of white muslin stretched over a wooden frame. The remaining three tanks were left unshaded, resulting in a total of 18 shaded and 18 unshaded pots.

The light intensity at the plant level as measured using a Skye SKP200 light meter was 245 $\mu E\ m^{-2}\ s^{-1}$ in the unshaded tanks and 38 $\mu E\ m^{-2}\ s^{-1}$.

The number of leaves on each plant was recorded at the beginning of the experiment of the experiment and again after a period of six weeks. Six plants from each treatment were harvested and the measurements described in section 2.2 were carried out.

After this six week period shading was removed from the treatment, so that all plants were in full light. After a further period of 17 weeks, plants were harvested and morphological measurements were carried out as before.

Section 3
RESULTS

3.1 Sediment Organic Content

Results for this experiment are summarised in Table 5.1. Figure 5.2 illustrates the allocation of biomass in *Littorella* plants grown in sediment of differing organic content. Analysis of variance showed a significant difference between the total biomass of plants grown at different sediment organic matter concentrations. Maximum biomass was observed in plants grown at 75% organic matter ($P = 0.024$). Shoot biomass also changed significantly with different organic matter content ($P = 0.047$).

Only plants grown in sediment with organic matter content of between 25 and 75% formed stolons, and the greatest stolon biomass occurred at a sediment organic content of 75% (Figure 5.2). Plants grown in 100% organic matter did not allocate resources to stolon development, but did have a significantly higher number of leaves than *Littorella* plants grown at any other sediment level (Figure 5.3).

Total biomass was not significantly correlated with sediment organic content. This is to be expected, as the response of *Littorella* to increased sediment organic content appears to follow a quadratic rather than linear relationship with optimal biomass occurring at intermediate sediment organic matter concentrations (see Figure 5.2).

Neither leaf area ratio (LAR) nor specific leaf area (SLA) showed any significant variation with sediment organic content (Figure 5.4). Total chlorophyll concentration also showed no variation with different sediment organic content, with a mean chlorophyll concentration of 14.6mg g^{-1} dry weight for plants grown in sediment concentrations ranging between 10 and 100% (Figure 5.5). Due to the small size of plants grown at 0% organic content, chlorophyll determination was not carried out.

Table 5.1 Summary of characteristics of Littorella plants grown with different sediment organic content

Organic Matter (% vol)	Biomass (mg)			
	Total	Root	Shoot	Stolon
	mean (s.e.)	mean (s.e.)	mean (s.e.)	mean (s.e.)
100	41.6 (3.4)	28.6 (2.9)	13.1 (0.9)	0.0 (n/a)
75	60.2 (19.6)	29.4 (6.7)	10.9 (2.2)	19.9 (9.6)
50	57.5 (26.8)	32.2 (4.0)	15.3 (2.8)	10.1 (8.1)
25	46.9 (10.3)	29.4 (5.6)	12.0 (3.1)	5.6 (5.6)
10	31.8 (7.6)	23.1 (5.0)	8.7 (2.5)	0.0 (n/a)
0	18.0 (2.8)	13.3 (2.5)	4.7 (0.6)	0.0 (n/a)

Organic Matter (% vol)	Leaf number mean (s.e.)	Chlorophyll content (mg/g dry weight) mean (s.e.)	Leaf area	
			LAR mean (s.e)	SLA mean (s.e.)
100	5.8 (0.25)	16.2 (1.6)	0.27 (0.06)	0.65 (0.04)
75	4.4 (0.26)	13.6 (0.6)	0.23 (0.04)	0.67 (0.08)
50	4.8 (0.37)	13.6 (1.8)	0.16 (0.01)	0.51 (0.05)
25	4.7 (0.47)	16.0 (2.6)	0.20 (0.03)	0.58 (0.07)
10	5.0 (0.41)	14.0 (0.4)	0.17 (0.01)	0.59 (0.03)
0	3.8 (0.37)	n/a	n/a	n/a

Organic Matter (% vol)	Leaf lengths (mm)	
	longest	average
	mean (s.e.)	mean (s.e.)
100	45.5 (3.9)	35.5 (2.5)
75	42.8 (6.3)	35.4 (2.6)
50	43.2 (4.2)	34.8 (2.1)
25	41.0 (4.9)	32.7 (2.3)
10	36.3 (6.9)	29.2 (3.0)
0	28.8 (6.8)	22.3 (2.8)

Figure 5.2 Effect of sediment organic content on Littorella biomass

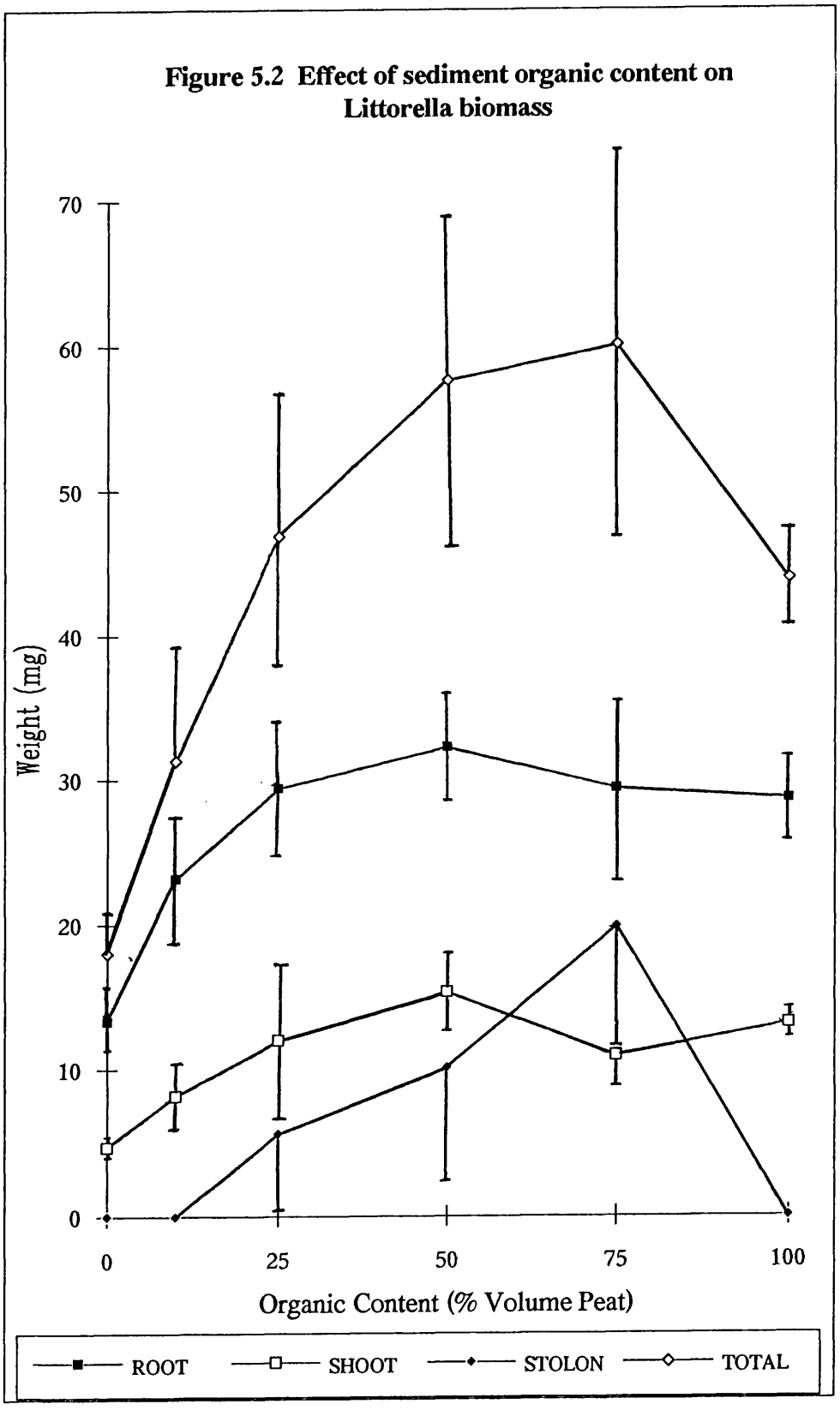


Figure 5.3 Number of leaves of Littorella plants grown with different levels of sediment organic matter

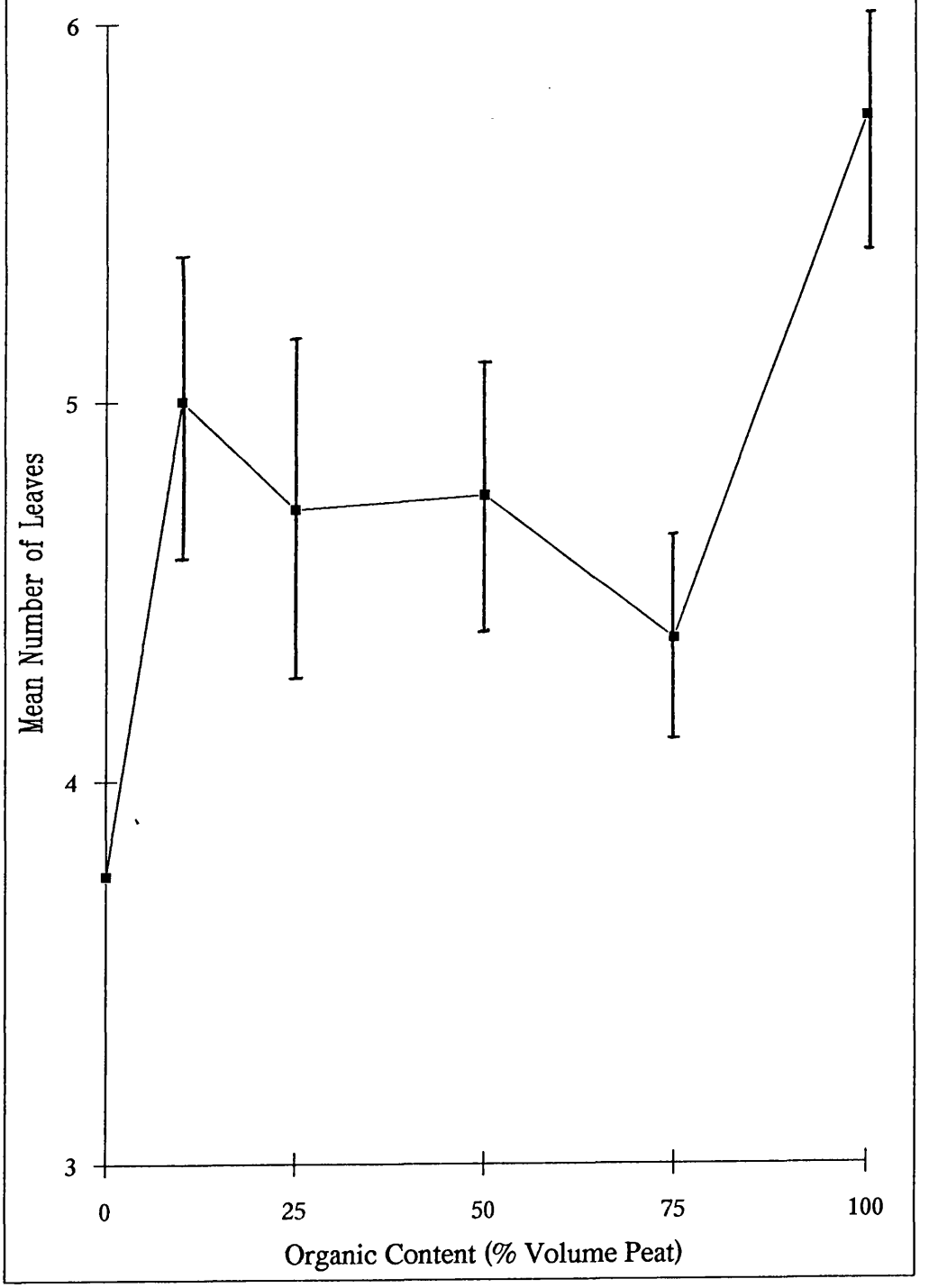


Figure 5.4 Effect of sediment organic content on Littorella leaf surface area

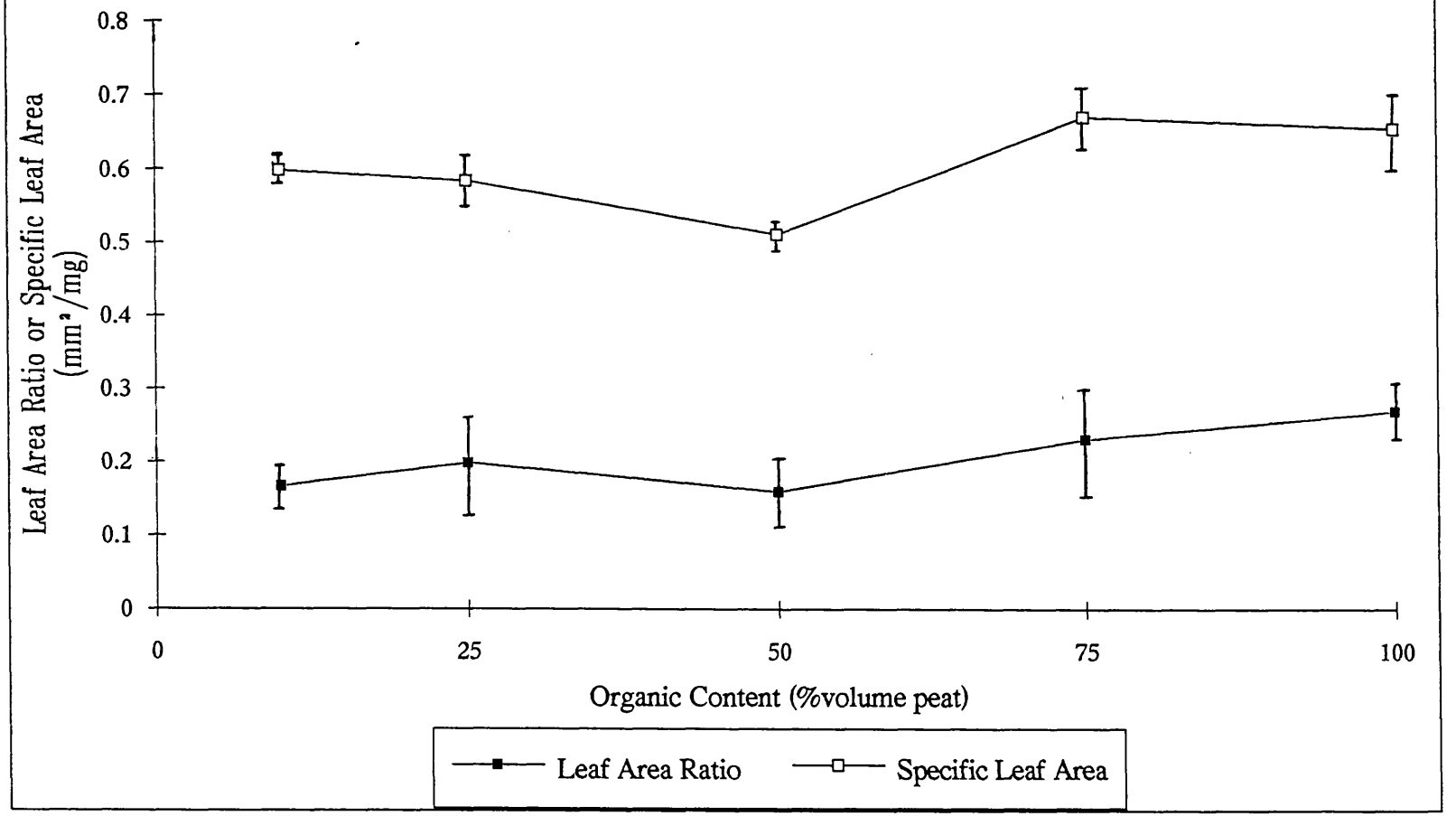
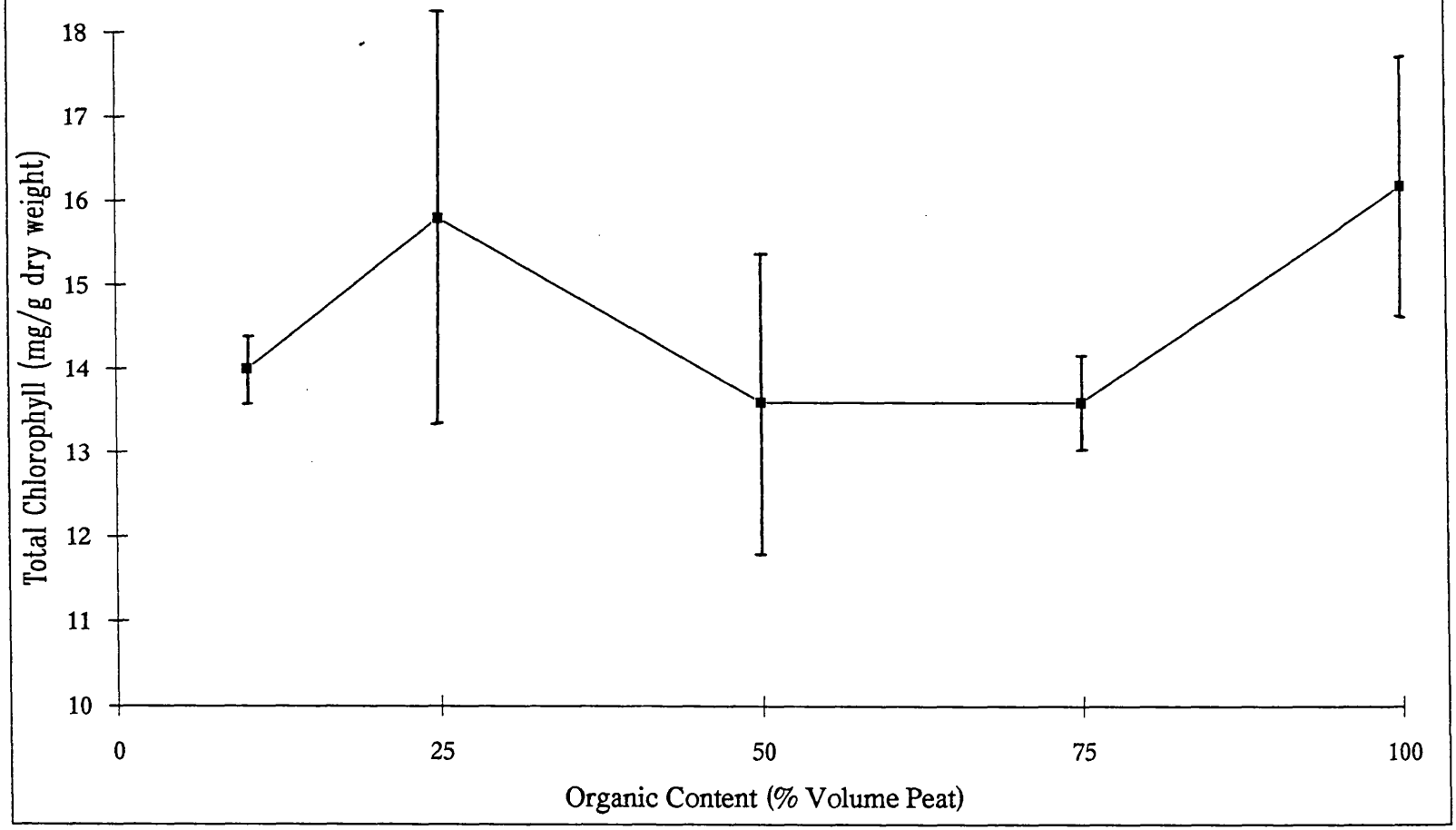


Figure 5.5 Effect of sediment organic content on Littorella leaf chlorophyll content



The optimum sediment organic concentration for *Littorella* growth under the conditions of this experiment was 75%, although any organic matter content greater than 25% was sufficient to allow stolon development and biomass accrual. All further greenhouse experimental work was carried out at a sediment organic content of 50%, as this peat concentration was easy to handle and produced good growth results. When a concentration of peat greater than 50% was used, the water became discoloured due to the release of humic substances.

3.2 Effects of Shading on *Littorella* Morphology and Leaf Chlorophyll Content

Plants that had been shaded for six weeks had a lower leaf biomass than unshaded plants (one way analysis of variance, $P = 0.03$). However there was no significant difference between the root biomass of the shade and control plants (Table 5.2; Figure 5.6).

Figure 5.7 shows the total numbers of leaves, stolons and new plants for the 6 control (light) and the total number of leaves for the 6 shade plants that were harvested after six weeks. Total leaf numbers quoted are for primary plants only and all offspring have been described as individual new plants. Neither stolons, nor new plants were formed by shaded *Littorella.*

A mean leaf accumulation of 2 leaves per plant throughout the 6 week experimental period was observed in unshaded plants. Total leaf number in the shade plants did not increase. However, by the end of the experimental period, three plants had lost one leaf, two plants had gained one and one plant showed no change in leaf number (Appendix E.3I)

These observations are supported by the fact that each unshaded plant had one small leaf, of length less than 2mm, whereas only three of the shaded plants had such small leaves. Plants that were rapidly accruing new leaves tended to have a greater number of small, newly developed leaves, whereas plants that were accumulating leaves more slowly tended to have fewer small leaves.

Table 5.2 Summary of characteristics of Littorella plants grown with and without shading

Organic Matter (% vol)	Biomass (mg)			
	Total	Root	Shoot	Stolon
	mean (s.e.)	mean (s.e.)	mean (s.e.)	mean (s.e.)
Light 1	33.3 (7.2)	15.3 (3.1)	15.7 (4.6)	2.3 (1.4)
Shade	14.1 (2.0)	9.8 (1.2)	4.2 (1.1)	0.0 (n/a)
Light 2	45.6 (7.8)	19.2 (3.4)	12.7 (4.0)	13.7 (7.9)
Shade Recover	37.5 (7.3)	17.4 (3.2)	14.3 (3.0)	6.6 (3.1)

Organic Matter (% vol)	Leaf number	Chlorophyll content (mg/g dry weight)	Leaf area	
			L.A.R.	S.L.A.
	mean (s.e.)	mean (s.e.)	mean (s.e.)	mean (s.e.)
Light 1	6.5 (0.50)	13.6 (1.8)	0.43 (0.07)	0.76 (0.06)
Shade	3.3 (0.49)	51.0 (6.8)	0.32 (0.04)	1.27 (0.02)
Light 2	5.2 (0.48)	18.2 (1.4)	0.29 (0.02)	0.87 (0.09)
Shade Recover	6.8 (0.65)	17.1 (1.54)	0.31 (0.03)	0.73 (0.05)

Organic Matter (% vol)	Leaf lengths (mm)	
	longest	average
	mean (s.e.)	mean (s.e.)
Light 1	40.2 (4.9)	35.1 (2.0)
Shade	38.0 (2.6)	31.2 (2.2)
Light 2	66.2 (6.4)	53.6 (3.0)
Shade Recover	59.7 (4.1)	39.5 (2.8)

Figure 5.6 Effect of shading and subsequent removal of shading on Littorella biomass

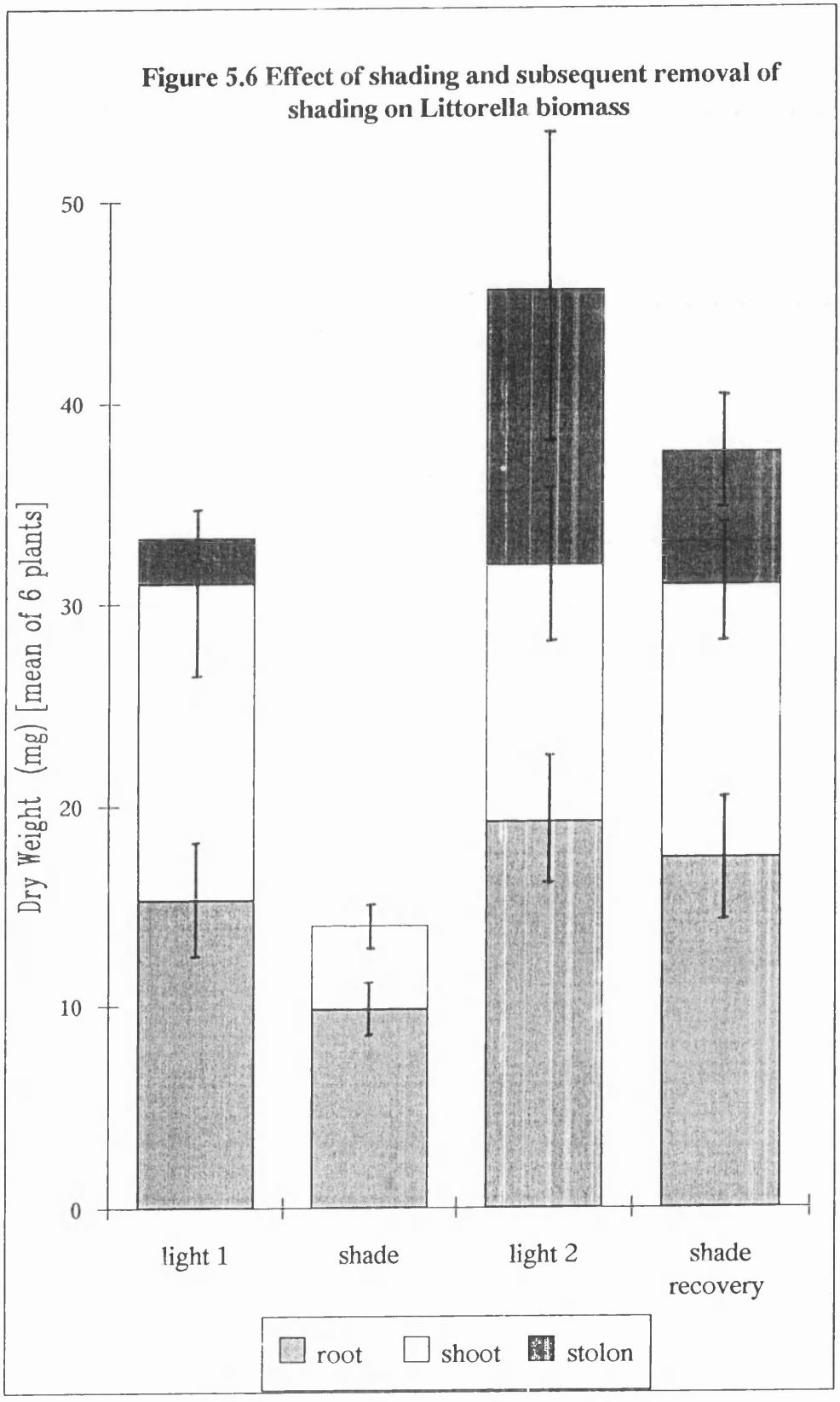


Figure 5.7 Effect of shading on Littorella leaf, stolon & number of new plants

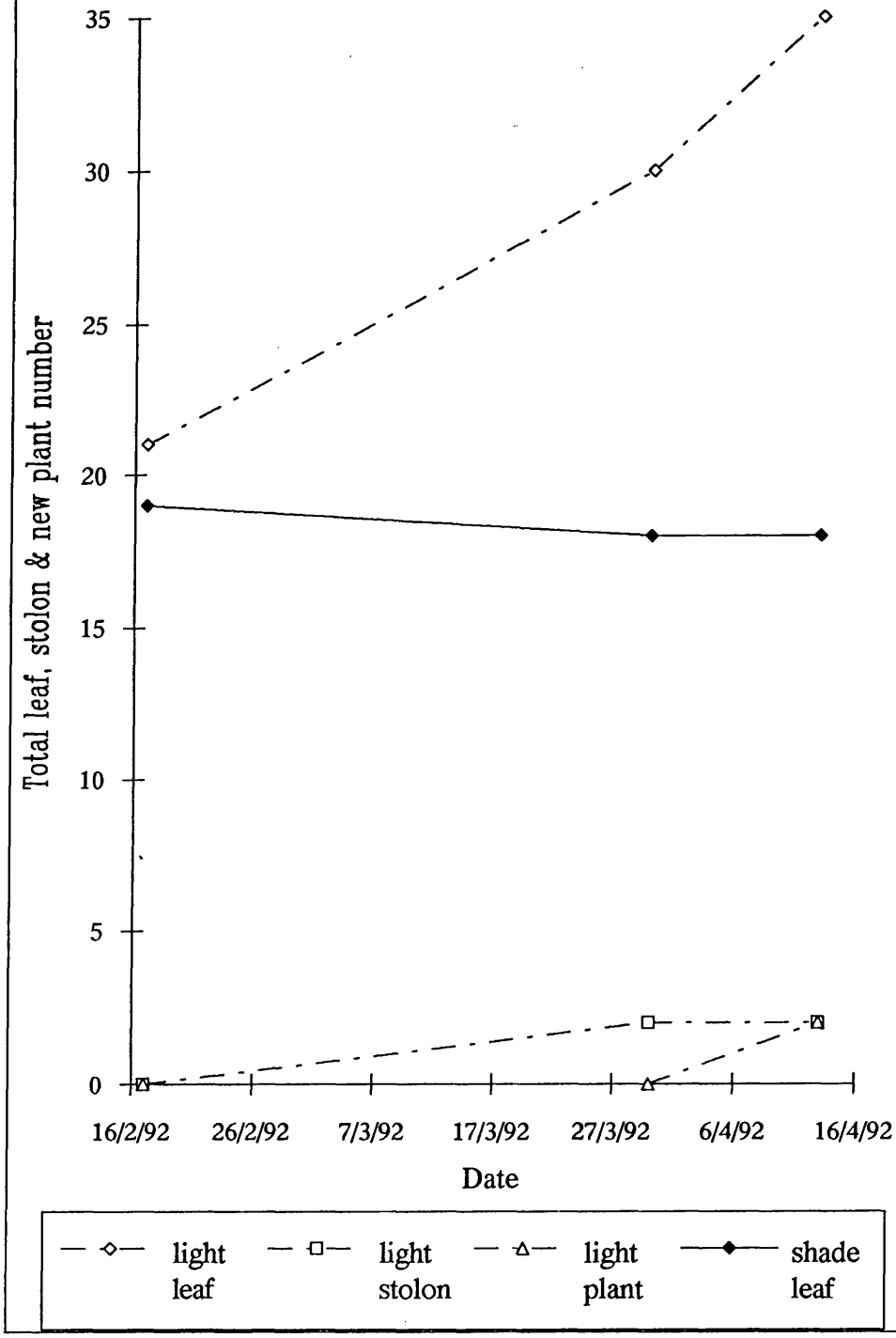


Figure 5.8 Effect of shading on chlorophyll content and chlorophyll a:b in Littorella

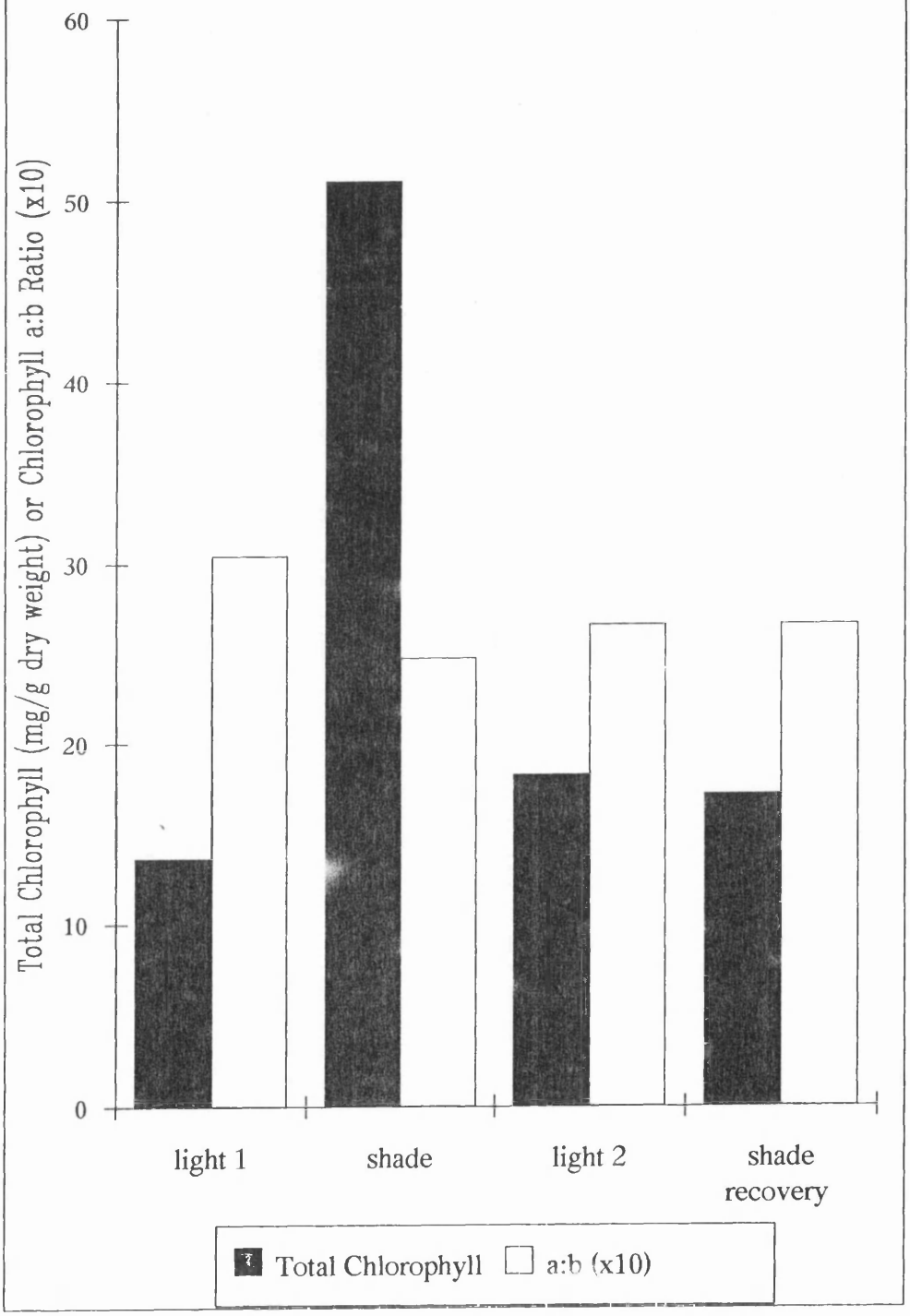
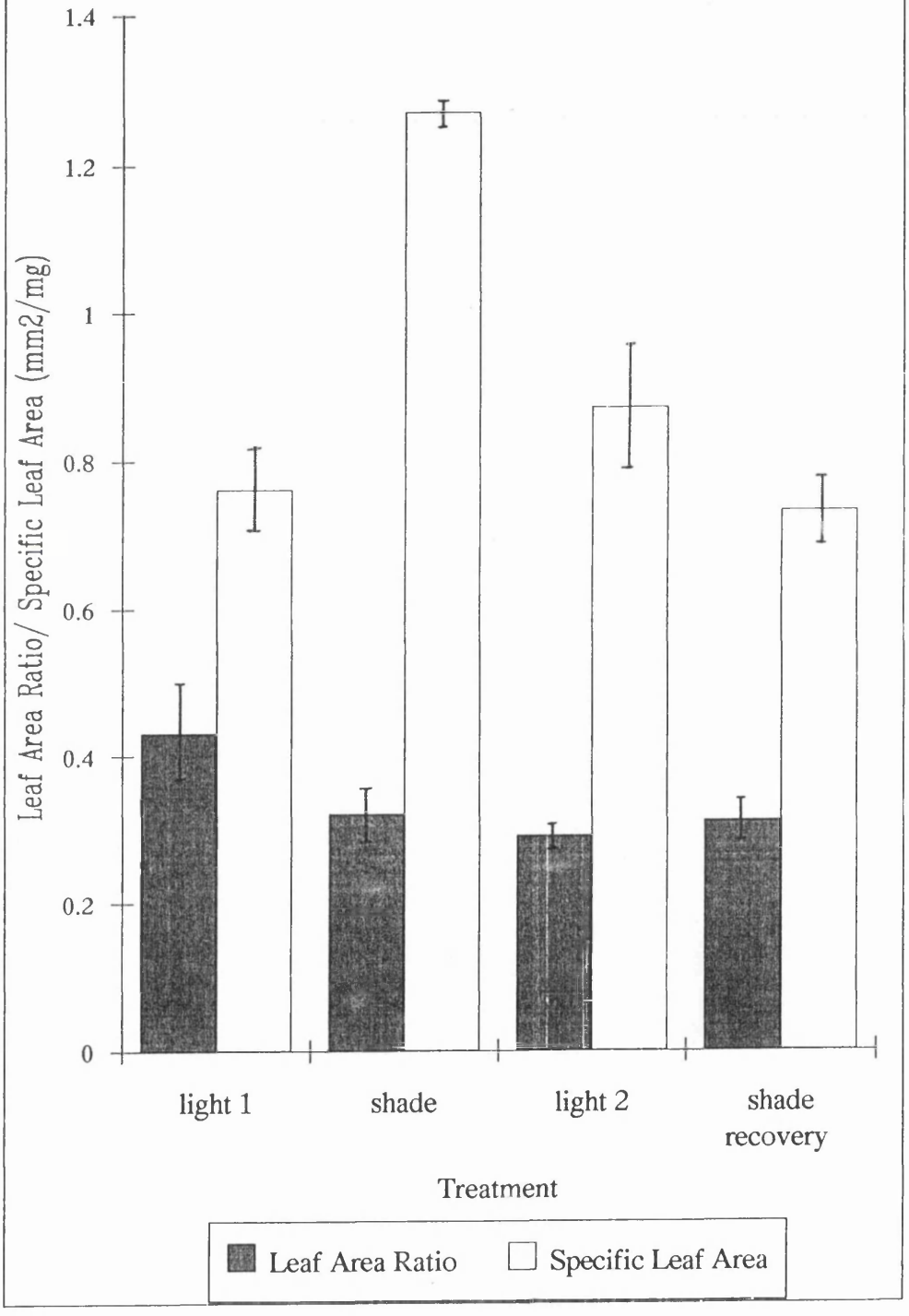


Figure 5.9 Effect of shading on Littorella leaf surface area



Total chlorophyll concentration in shaded leaves was significantly higher than that of unshaded leaves (one way analysis of variance P = 0.007) with mean chlorophyll content of shaded and unshaded leaves being 51.0 and 13.6mg g^{-1} dry weight respectively (Figure 5.8). Chlorophyll *a:b* was lower in shaded plants.

There was no significant difference between the LAR of shaded and unshaded plants; however the SLA was significantly higher (P = 0.033) in shaded plants (Figure 5.9). No change in the LAR can be attributed to the fact that there were no differences between the root biomass in the control and shaded plants.

3.3 Effect of Shading and Subsequent Removal of Shading on *Littorella* Morphology and Leaf Chlorophyll Content

Figure 5.10 shows the effect of shading and subsequent removal of shading (time of shading removal depicted by arrow) on *Littorella* total number of leaves, stolons and new plants. Data are presented for the total number of leaves, stolons and new plants of the 6 shaded and the 6 unshaded control plants (referred to as recover and light 2 respectively).

Prior to the removal of shading, shaded and unshaded plants followed the same biomass accumulation pattern as described in Section 3.2, with no significant net increase being observed in the shade plants and an increase in both total leaf number and stolons in the unshaded plants (Figure 5.10).

Three weeks after the removal of shading there was an increase in the total number of leaves of previously shaded plants (Figure 5.10). Total leaf number in the unshaded plants at this stage showed a slight decline that coincided with the beginning of new plant formation. During this period, there was no gross increase in leaf number of the mature plants and four plants lost one leaf each (Appendix E2.I).

Figure 5.10 Effect of shading and subsequent removal on Littorella leaf, stolon & new plant production

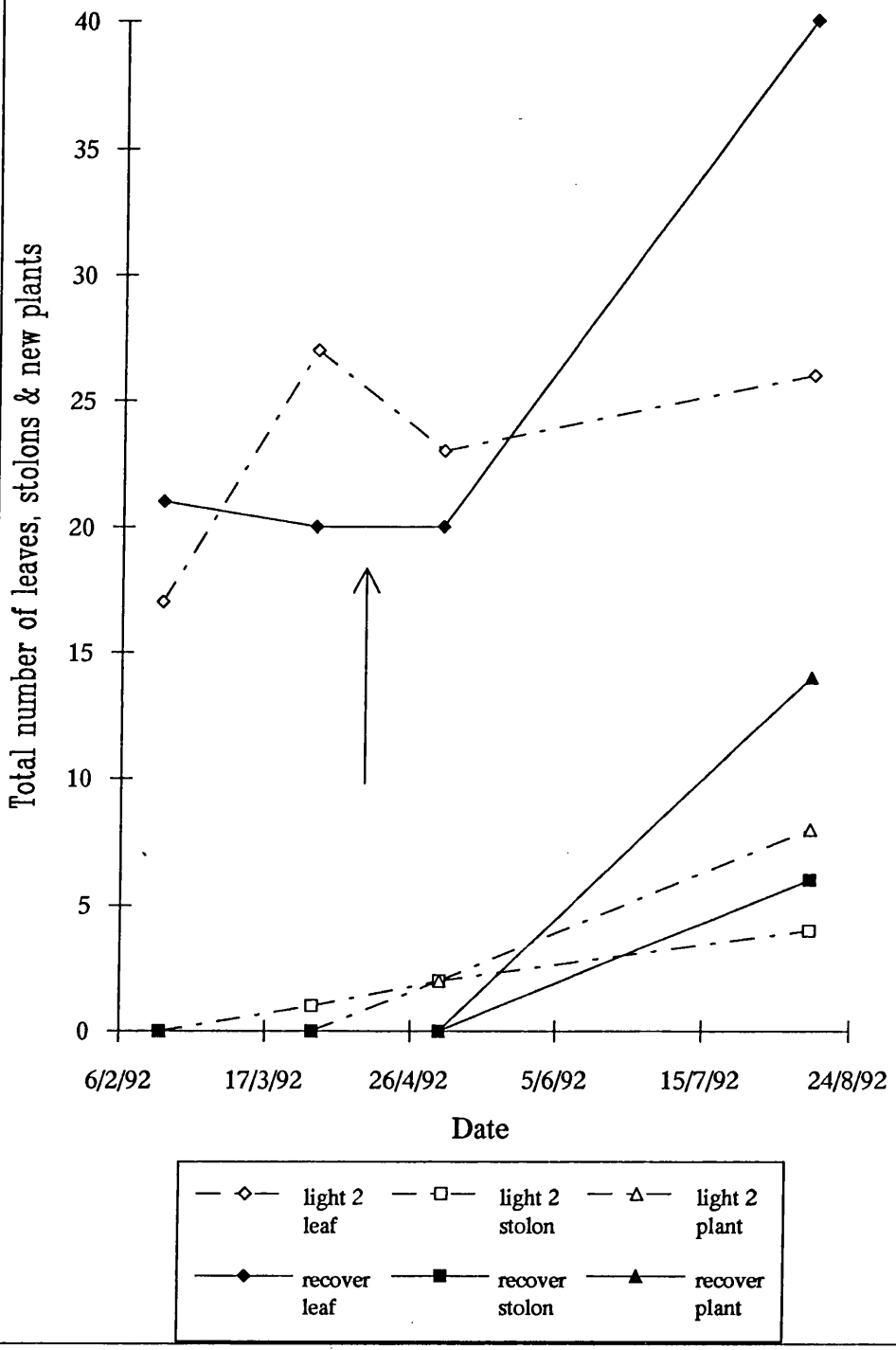
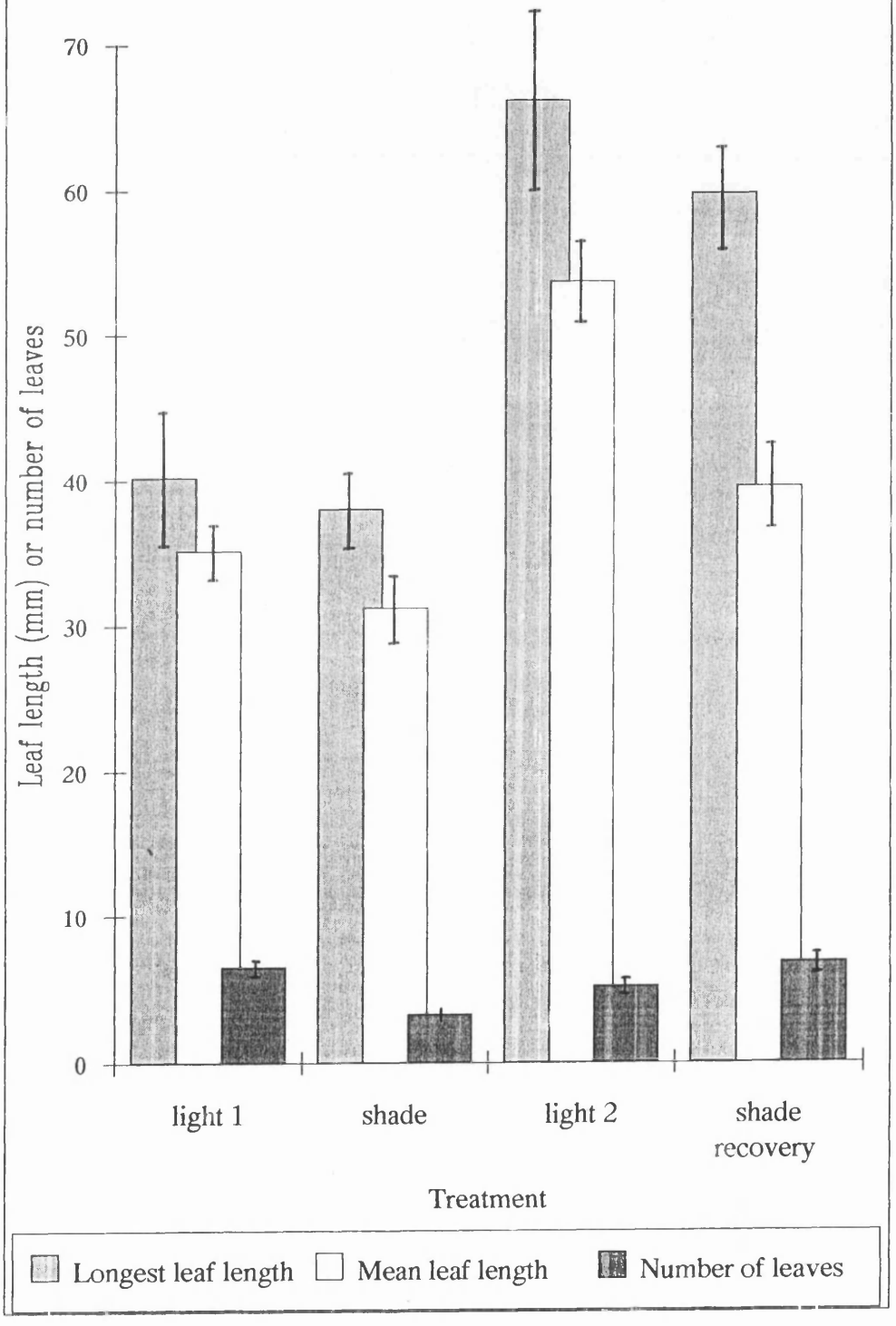


Figure 5.11 Effect of shading on Littorella leaf length and number of leaves



Seventeen weeks after the removal of shading, total chlorophyll content and chlorophyll *a:b* ratio of previously shaded *Littorella* plants did not significantly differ from those of unshaded plants (Figure 5.8). There was also no significant difference between LAR and SLA in previously shaded and unshaded plants (Figure 5.9).

Seventeen weeks after the removal of shade, shaded plants doubled the total number of leaves, whereas the total leaf number of the unshaded control plants remained constant (Figure 5.10). There was no significant difference between the mean leaf biomass of unshaded controls and shaded plants (Figure 5.6); the plants that had been shaded produced many small leaves in comparison with plants that had not been shaded. There was no significant difference between new plant biomass of unshaded control plants and shaded plants (Figure 5.6). The shaded plants, in total, produced 14 new plants. Unshaded plants, however, only produced a total of 8 new plants throughout the duration of the experiment (Figure 5.10). Shade recovery plants had a greater number of smaller new plants, 17 weeks afer the removal of shading, when compared with plants that had not been shaded.

Seventeen weeks after shade removal, analysis of the longest leaf data revealed there to be no significant difference between shaded and unshaded control plants ($P = 0.371$). Many of these leaves were among the three oldest leaves that had been produced prior to shade application. There was also no significant difference between the mean leaf length of treated and untreated plants ($P = 0.57$) prior to the removal of shading (Figure 5.11). Seventeen weeks weeks after the removal of shading, mean leaf length of unshaded controls were significantly longer than those of shaded plants ($P = 0.001$). Youngest leaves were omitted from this analysis as their inclusion would increase variation in the data set and bias the results towards shorter leaf lengths. This would not reflect the true leaf length, as the young leaves had not grown to their mature size.

Section 4
DISCUSSION

Misra (1938) noted the absence of macrophytes in areas with high organic content and related this to high hydrogen ion or low metal ion concentration. Smits *et al.* (1990), studied the root oxygen leakage patterns and alcohol fermentation ability of several aquatic plants found typically in a range of different trophic habitats. These workers hypothesised that *Littorella* did not survive well in sediment of high organic content due to a poorly developed root aerenchyma and high root oxygen permeability compared with species such as *Nuphar* and *Nymphea.* Farmer & Spence (1986), however, noted that *Littorella* could successfully colonise pure peat in Scottish lochs.

A non-linear growth response to increased sediment organic content has been observed in other species of isoetid, e.g. *Lobelia dortmanna* (Wilson, 1991).

In a lake with low sediment organic content (0.3 - 5.1% w/w) Sand-Jensen & Søndergaard (1979) observed a positive significant regression between *Littorella* biomass and shoot weight with sediment organic content at a depth of 0.75m. This relationship was not observed in plants from deeper water, where low irradiance was considered to be an important determinant.

Under conditions of low light intensity, *Littorella* did not produce new leaves, but an increase in the total leaf chlorophyll, and a decrease in the chlorophyll *a:b* ratio, was observed. Seventeen weeks after the removal of shading, leaf chlorophyll concentrations in previously shaded plants were the same as in unshaded plants. Holstrup & Weigleb (1991b) noted that *Littorella* under conditions of low light showed no morphological changes. Shade plants had a higher SLA than unshaded plants indicating that the area of leaf in relation to leaf biomass was greater in shaded plants. As there was no new leaf accumulation in the shade plants, and the unshaded plants had been grown in the same pre- and post-shade light regime, it would appear that the shaded leaves had adapted morphologically to the reduced light levels by increasing their surface area relative to the leaf biomass. After the

removal of shading the SLA of shaded plants became the same as that of unshaded plants.

An increase in the SLA of macrophytes in response to shading has been well documented (e.g. Spence & Chrystal, 1970b). Spence *et al.* (1973) correlated an increase in SLA in *Potamogeton obtusifolius* throughout the growing season with phytoplankton growth. In the same study, no changes in SLA were observed in plants sampled from lochs that did not experience algal blooms. In contrast, Søndergaard & Bonde (1988), reported there was no significant difference between the SLA of plants sampled from depths of 0.2m and 2.3m. These workers stated that *Littorella* has a very rigid leaf structure that prevents significant changes in SLA.

Seventeen weeks after the removal of shading, *Littorella* plants appeared unaffected by their history of low light levels in terms of biomass as there was no significant difference in the relative biomass allocation to roots, shoots or stolons (Figure 5.6). However, plants that had experienced shading had a greater number of smaller leaves and had produced more new plants, than those that were unshaded. In Chapter 4, stolon production per plant was positively related to filamentous algal biomass. From field populations, it was not possible to determine the numbers of new plants that were produced per primary plant, but as each stolon will give rise to at least one new plant, stolon production can give an indication of new plant production (i.e. a greater number of stolons will give rise to greater number of new plants).

Woodward (1990) states that in woodland plants, shade-tolerant plants will form a denser canopy and so limit species diversity by preventing seedling development. The development of a greater number of leaves and new plants in *Littorella* that have experienced shading may be a similar response that may reduce competition in a stressed environment.

The increase in leaf number and new plant production that follows transfer of plants from deep shade conditions may have a range of implications for the

ecology of *Littorella* in Scottish lochs. The experimental plants in this work were all obtained from a site in Loch Lomond that did not experience algal mat formation, phytoplankton blooms or have high level of epiphytic colonisation, and the response of plants, with different genotypes, located at other sites may be quite different. Robe & Griffiths (1992) attributed differences in ramet production in *Littorella* plants sampled from two different lakes to genetic diversity.

Section 5
CONCLUSION

Littorella demonstrates a quadratic growth response to increased sediment organic matter content, with maximum growth occurring at intermediate concentrations of sediment organic matter.

Severe shading of *Littorella* can be tolerated for a period of several weeks, during which time there is an increase in leaf chlorophyll, a decrease in chlorophyll *a:b*, a cessation of new leaf development and an increase in SLA. During the shade period no significant net increase in biomass occurs. After the removal of shading the growth of previously shaded plants is more rapid when compared to unshaded controls. Consequently, after a period of 17 weeks there was no difference between the total biomass and biomass allocation in shaded and unshaded plants. Shaded *Littorella* plants grow a greater number of smaller leaves and produce more new plants than unshaded plants when returned to high light.

Chapter 6:

Adaptation of *Littorella* to Shade - A Further Greenhouse Study

Chapter 6:

Adaptation of *Littorella uniflora* to Shade: A Further Greenhouse Study

Section 1
INTRODUCTION

Field observations in Chapter 4 provided evidence of physiological changes in *Littorella* that occurred at the same time as filamentous algal mat formation. These changes in chlorophyll and leaf nitrogen content, suggested a shade adaptation. In Chapter 5, data were presented which discussed morphological adaptations to long term shading that may be brought about by the presence of a filamentous algal mat growing above *Littorella* in the water column.

One morphological trait that confers an advantage to aquatic macrophytes in low light regimes is a high biomass near the water surface (e.g. *Myriophyllum spicatum,* Titus *et al.*, 1975). Plants in low-irradiance environments that lack this morphology, such as *Littorella*, will need to develop physiologically in order to grow and succeed in this type of environment.

Titus & Adams (1979) considered mechanisms whereby *Vallisneria americana* could overcome its less-than-ideal morphology. Their work showed that this species was physiologically more adaptable to low light levels than *M. spicatum.* Spence & Chrystal (1970a,b) showed that plasticity in morphology and physiology was important to the depth distribution of species of *Potamogeton.*

This chapter reports the physiological adaptability of *Littorella* to shading. The first experiment described considers the rate of adaptation of *Littorella* to shading by measuring the change in leaf chlorophyll levels with time after the application of shading. The second experiment considers changes in the photosynthetic light curves of *Littorella* and measurements of $\Delta^{13}C$.

Section 2

METHODS

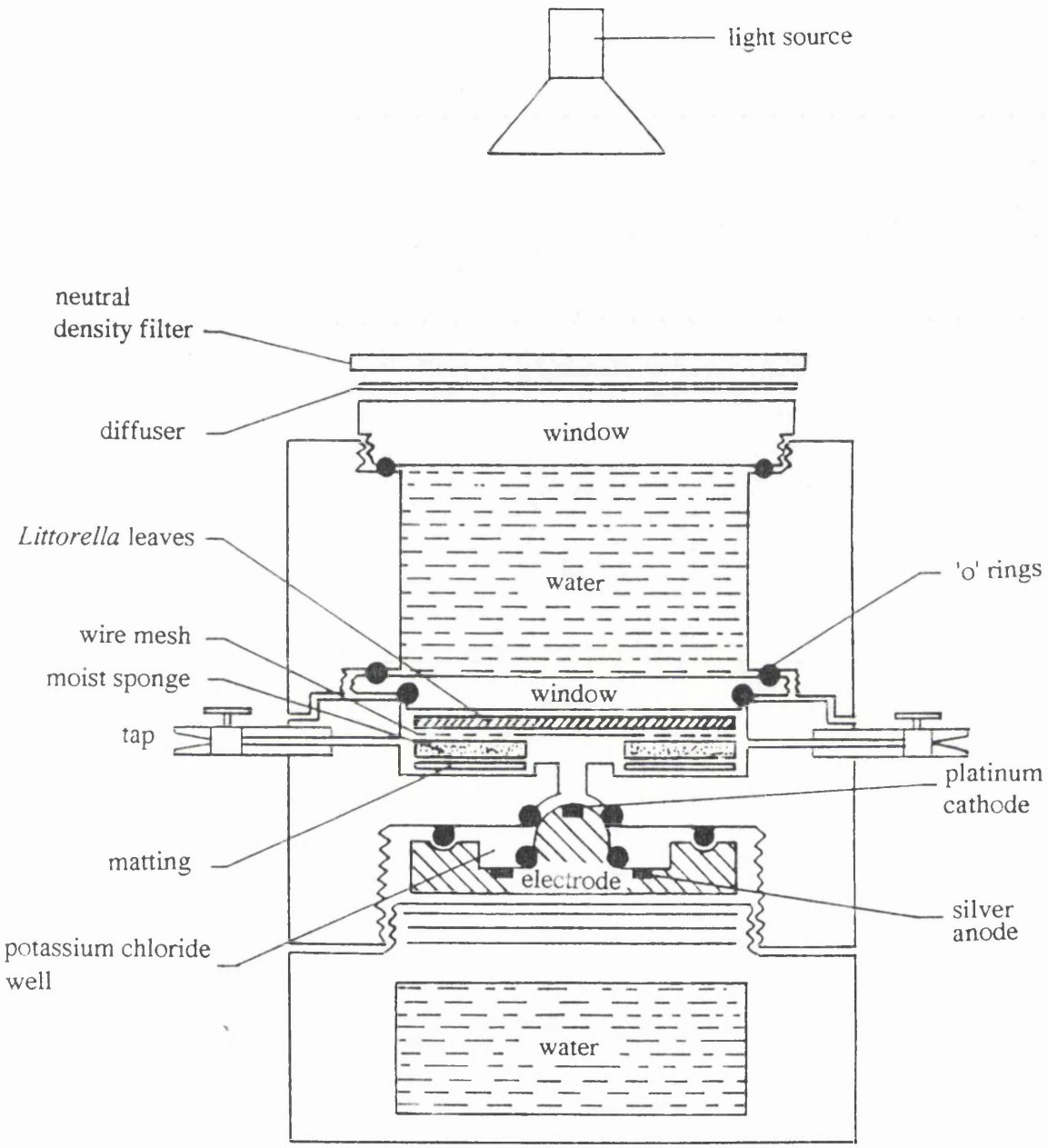
2.1 Rate of Response of *Littorella* to Shading

Littorella plants were planted in a 36cm seed tray in a 50:50 peat:sand sediment mix. Plants were grown in the greenhouse for a period of six months in natural daylight supplemented with mercury lamps at an irradiance of approximately 400 $\mu E\ m^{-2}\ s^{-1}$. After this period the plants had developed into a sward, similar to that observed under field conditions (approximate plant density = 2,000 m^{-2}). Two trays were shaded using the same shading regimes described in Chapter 5, and two trays were left unshaded. Chlorophyll content was measured as described in Chapter 4, prior to and 2, 3, 5, 9, & 11 days after the application of shading. Chlorophyll content was also measured in the untreated plants at the start of the experiment and on days 2, 5, 9 & 11. Two plants were taken from each tank, resulting in four samples per measurement per treatment. The results for time point were pooled for data analysis

2.2 Determination of Photosynthetic Efficiency

Lipkin *et al.* (1986) studied three methods of measuring photosynthesis rates (changes in O_2 production, $^{12}CO_2$ uptake from the medium and ^{14}C uptake by plants) and found no reason to favour any one method. In the experiment reported here, photosynthetic efficiency was calculated using measurements of oxygen evolution at different light intensities. The measurement of oxygen evolution with time allows continuous measurement of photosynthesis (Walker, 1990; 1993) so any lag phase in photosynthesis after the application of such a treatment may be observed (Westlake, 1978).

Figure 6.1 Schematic Diagram of a Gas Phase Oxygen Electrode



2.2a Determination of Light Response Curves.

Information on use of the leaf electrode is outlined in Walker (1993) and full operational details are presented in Walker (1990). The electrode was set up as shown in Figure 6.1 with half-saturated potassium chloride (KCl) solution in the well. Half-saturated KCl evaporated more slowly than fully-saturated KCl so extended the useful working life an electrode. Prior to setting up the electrode, nitrogen gas was bubbled through the KCl in order to remove any oxygen present and so reduce the time taken for the electrode to give a steady reading.

Carbon dioxide was supplied in the form of 200µl of 1 molar potassium carbonate buffer. The was applied to a capillary mesh sandwiched between two stainless steel wire discs in order to prevent both the plant material and the electrode from coming into direct contact with the alkaline buffer solution. A second capillary mat moistened with tap water was placed between the wire mesh and the leaf chamber in order to raise the relative humidity inside the leaf chamber and prevent the leaves under test from dehydrating.

Preliminary studies with the oxygen leaf electrode were carried out with intact *Littorella* leaves. Intact leaves showed a slow response to changes in light intensity (measured by increase in oxygen evolution).

Westlake (1978) presented data for lag phases of intact leaves with lacunae and suggested that measurement of oxygen evolution can be carried out as long as a steady state is reached prior to taking any readings. However, in the case of *Littorella,* which have extensive lacunae compared to the species used by Westlake (*Myriophyllum spicatum* and *Vallisneria americana*), lag times are so long that the measurement of light response curves is impractical.

In order to obtain measurements of change in oxygen evolution leaves were sliced into two halves vertically using a new sharp razor blade. The cut surface was placed on the moist capillary matting and the cuticle side containing the chloroplasts was orientated towards the irradiance source. All the leaves from one

plant (n = 4) were used in each determination and were arranged in the chamber, so that no overlapping occurred.

The use of all the leaves in a plant overcomes the problems in differences in respiration rate and photosynthetic efficiencies of old and young leaves (e.g. Adams *et al.* 1974), although Madsen (1987b) reported there to be no physiological differences between old and young leaves in *Littorella.* As entire plants were not used the results obtained here cannot be extrapolated to field conditions since no account has been taken of other tissues such as roots or stolons (Hootsmans & Vermaat, 1991; Azcon-Bieto, 1992).

Temperature was maintained at 25°C using a circulator that was cooled by running tap water. This temperature was selected as this was similar to the temperature maintained in the greenhouse and it was possible to maintain the electrodes at this temperature without the use of refrigeration. After the electrode recorded a constant oxygen concentration and before any measurements were carried out, each electrode was calibrated by flushing the system out with nitrogen - full calibration details can be obtained in Walker (1990, 1993).

Irradiance was supplied by 24°, 50W dichroic quartz-halogen spot lamps (Wotan) with a diffuser (tracing paper) placed between the light source and the leaf chamber window in order to insure an even light distribution over the sample was obtained. One lamp was used per leaf electrode. Different light intensities were obtained by placing a neutral density filter (Balzar, Lichenstein) between the light source and the leaf chamber window. Measurements were started on dark adapted tissue, to obtain a dark respiration rate, then irradiance increased in 13 increments up to approximately 550μE m-2 s-1.

2.2b Selection of a Model of Photosynthesis

In order to carry out statistical analysis, data obtained from light response curves must be obtained objectively (Sokal & Rohlf, 1981). As a consequence of this, data derived from hand drawn curves or by carrying out linear regressions on the light-limited section of the curve with a subjectively selected photosynthetic

maximum are not acceptable (Hootsmans & Vermaat, 1991). Several models have been used to describe photosynthetic light response curves and are reviewed by Jassby & Platt (1976).

Hootsmans & Vermaat (1991) compared the rectangular hyperbola (Michaelis-Menten) model and the hyperbolic tangent in their study of *Potamogeton pectinatus*. They demonstrated that there was no difference between the two models under conditions where photosynthesis was not inhibited by light at high light intensity. Under conditions of inhibiting high light intensity the Michaelis-Menten model breaks down (Duncan *et al.*, 1967). In the experimental set-up described above, the conditions required for the application of the Michaelis-Menten model were upheld.

The rectangular hyperbola model is described by the following equation:

$$P = \frac{Pmax\ I}{I + k}$$

Where P = Gross photosynthesis
Pmax = asymptotic rate of photosynthesis
I = light intensity
k = constant where I = 1/2 Pmax

Full details of its computation are presented in Duncan *et al.* (1967).

2.2b Carbon Isotope Discrimination

Most elements in the environment have more than one form of stable isotope. One form however is usually considerably more abundant than any other. Carbon has two stable isotopes, ^{12}C and ^{13}C which, in terrestrial systems, have an average abundance of 98.89% and 1.11% respectively (Ehleringer & Osmond, 1989). Variation in the level of stable isotopes can be used in a wide range of ecological studies such as photosynthetic pathway determination (e.g. Tenhunen *et al.*, 1982) and defining animal food sources (e.g. Stephenson & Lyon, 1982).

Isotope concentration can be measured using a mass spectrometer and the results obtained compared to that of a standard using the following equation

$$\Delta Xstd = (Rsam/Rstd-1)1000$$

where Xstd = isotope ratio in delta units relative to a standard.
Rsam = isotope abundance ratio of sample
Rstd = isotope abundance ratio of standard

Results are expressed as parts per thousand (‰). The standard used in carbon isotope discrimination is PeeDee Belemnite (a limestone) and results are expressed on the PBD scale (Ehleringer & Osmond, 1989).

The use of stable carbon isotopes in the study of C4 and C3 photosynthetic pathways is based on the differential use of stable isotopes by phosphenolpyruvate (PEP) carboxylase and ribulose bisphosphate (RuBP) carboxylase, the primary carboxylating enzymes of C4 and C3 photosynthesis respectively (Farquhar *et al.*, 1989). $\Delta^{13}C$ values range from -9 to -14‰ and from -20 to -35‰ for C4 and C3 plants respectively. In plants that carry out Crassulacean Acid Metabolism (CAM) the range of $\Delta^{13}C$ values is greater (-9 to -32‰), depending on the relative magnitudes of the PEP carboxylase and RuBP carboxylase reactions (Ehleringer & Rundel, 1988).

Changes in $\Delta^{13}C$ have been observed in forest canopies. Plants sampled lower in the canopy had a lower $\Delta^{13}C$ than those higher up. Interpretation of such results has provided controversy due to the difficulties in separating the effects of changes in irradiance and differences in source carbon dioxide (Farquhar et al., 1989). To the author's knowledge no study on the effects of shading on $\Delta^{13}C$ in submersed aquatic macrophytes has been carried out to date.

2.2d $\Delta^{13}C$ Measurements

The entire *Littorella* leaf biomass was harvested from each tank, washed in distilled water and dried at 60°C for 24 hours in an oven. The dried leaf material was then

coarsely ground in a coffee grinder. All further work was carried out at the Invergowrie Crop Research Institute, Dundee.

The dried samples were ground further in a Ketsch Ball Mill for 5 minutes until the plant material was the consistency of corn flour. Fine grinding of samples is essential in order to ensure thorough mixing of pooled samples and to ensure even burning in the combustion unit (Ehlringer & Osmond, 1989).

Approximately 1mg of each air-dried sample was accurately (2 decimal places) weighed into a separate tin cup. The tin cup was then crimped into a tight ball and placed in a labelled container. Two replicates of each sample were measured. $\Delta^{13}C$ measurements were carried out using continuous flow ANCA-MS on a Europa Scientific Tracermass System - the process is outlined below.

The loaded cups were placed into an automated sampler, which sent the samples to an automated combustion unit. After purification the resultant gases were sent to a mass spectrometer for analysis. Results were calibrated to a concentration of $\Delta^{13}C$ in PeeDee Belemnite in accordance with international standards using computer software. The system also measured total nitrogen and total carbon content. This method routinely has a variation of <0.3%. and is described in detail in Handley *et al.*, 1993.

Results from the three tanks in each treatment were pooled for data analysis.

2.2e Chlorophyll Concentration

Two plants were harvested from each tank (6 plants per treatment; total = 18) and the total chlorophyll levels and chlorophyll *a:b* were measured as described in Chapter 4.

2.2f Experimental Set-up

In most published work on shading experiments, PAR is reduced by the application of neutrally absorptive shading material (e.g. Barko & Filbin, 1983; Hootsmans &

Vermaat, 1992; Spink, 1993). As reported in Chapter 5, a reduction in PAR results in a decrease in chlorophyll *a:b* in *Littorella*. Chlorophyll *b* is associated with the light-harvesting complex chlorophyll *ab*-protein and which is associated with photosystem II (Björkman, 1981).

Chambers & Spence (1984) observed a decrease in red light (660nm) in relation to far red light (730nm) with increased depth and with increased phytoplankton chlorophyll *a* in Scottish lochs. Far red light is primarily utilised in photosystem I. Under conditions of far red light enrichment (e.g. under an algal mat) an increase in photosystem II (and consequently chlorophyll *b*) would result if a more equal balance in electron transport between the two systems is to be achieved (Björkman, 1981).

Changes in the proportion of red light in relation to far red light would not be brought about by the use of neutral density shading materials. In order to mimic the quantitative and qualitative change in light in natural systems, a method of algal shading was devised and the results compared to those obtained from neutral-density shading material that reduced PAR to a similar extent. To be able to study the effects of algal shading in the absence of other parameters, such as carbon and nutrient competition, or possible algal allelopathy (Van Vierssen & Prins, 1985), it was necessary to culture the algae in separate containers above the *Littorella* tanks.

Filamentous algae were not used for this experiment due to difficulties in obtaining a culture of uniform density across the surface of the tray. The possibility of using a phytoplankton culture was discounted due to rapid doubling times leading to difficulties in maintaining constant shade conditions throughout the duration of the experiment.

Ulva lactuca is a marine macroalga that commonly occurs on rocks, in pools and on other algae in the littoral and sublittoral zones of the sea. This species has broad roughly circular thalli ranging from 1cm up to 1m in size (Burrows, 1987). The flat sheet-like structure of the thallus makes it possible to layer plants on top of one-another in order to achieve the desired shading effect - it was possible to obtain a uniformly shaded environment for the *Littorella* to be cultured under.

Figure 6.2 Comparison of transmission spectra for filamentous algae (hatched line) collected from Loch Dee on 13th August 1991 and *Ulva lactuca* (solid line) used in shading experiment.

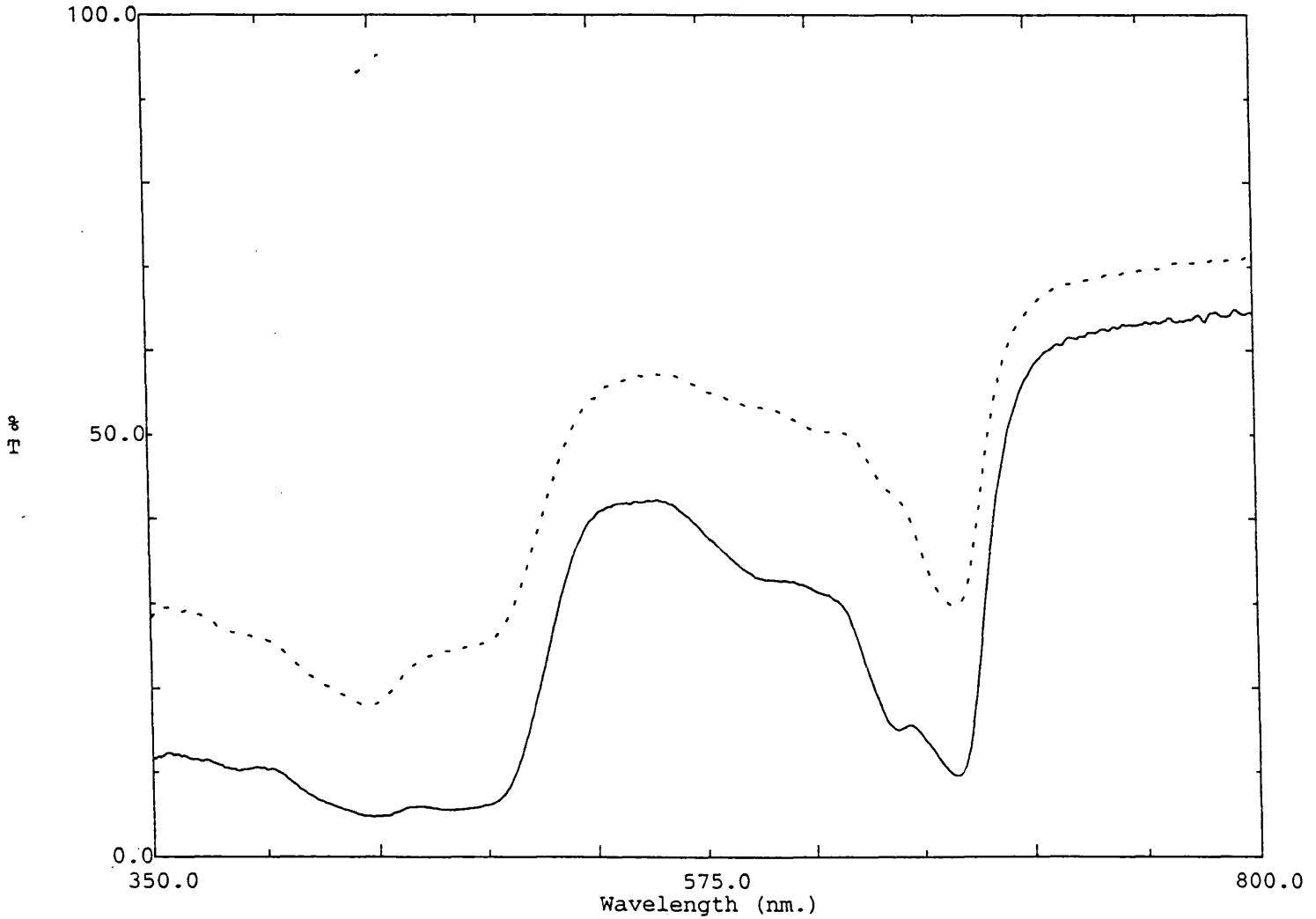
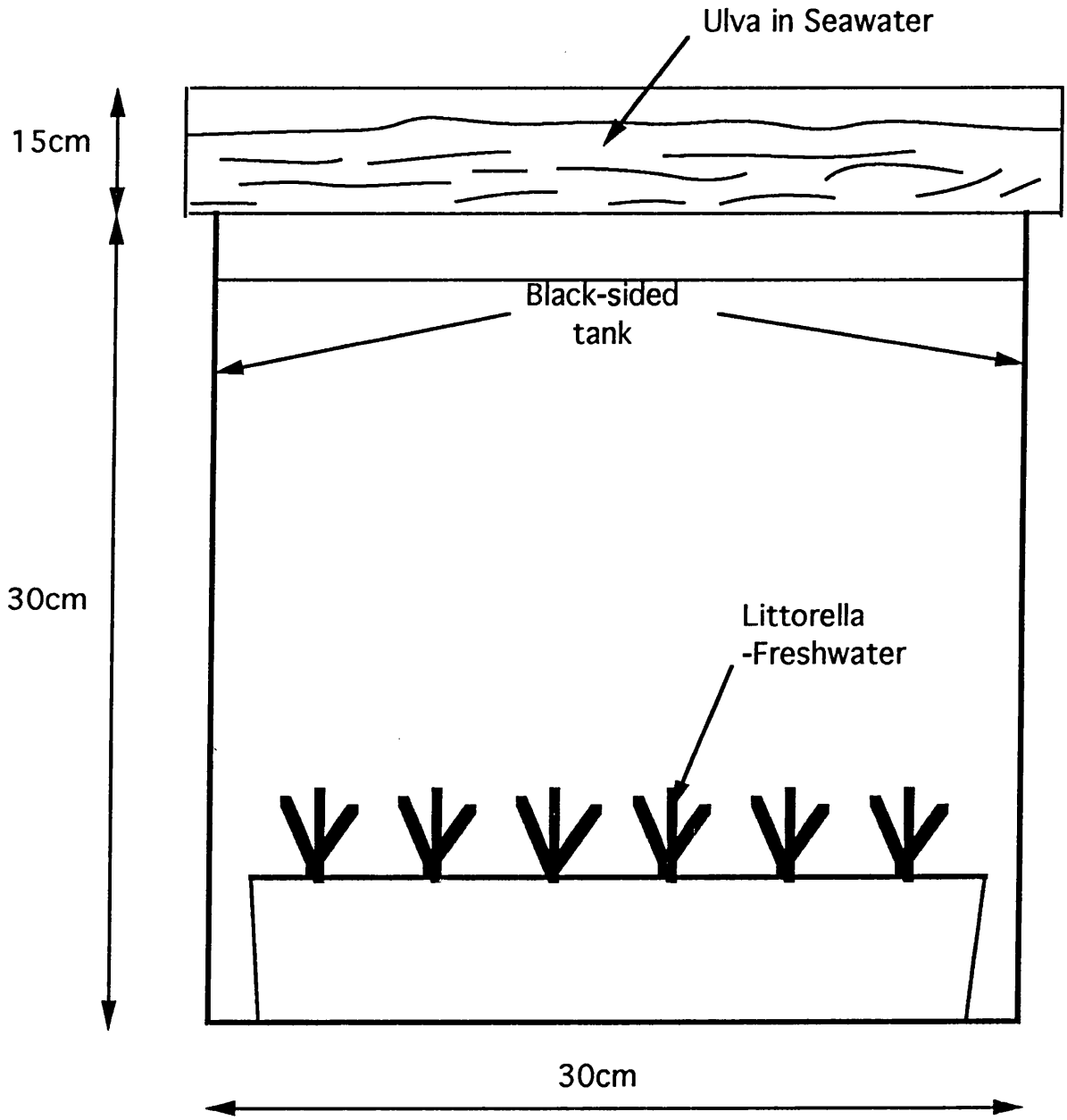


Figure 6.3 Schematic Diagram of Algal Shading Apparatus.



Although a marine species, *Ulva* has a similar transmission spectrum to filamentous algae collected from Loch Dee (Figure 6.2) and consequently was a useful shade model for the purpose of this work. The shading system used in this experiment is depicted in Figure 6.3.

Littorella plants were planted in 50:50 peat:sand in 36cm seed trays and were allowed to grow until a sward similar to that observed in field situations developed (ca. 2,000 individuals per m^2 approximately six months after planting) Plants were cultured in the circulatory system described in Chapter 5.

Three tanks were shaded with *Ulva* (Figure 6.3); three were shaded with the same white muslin as utilised in Chapter 5; and three were left unshaded (total: 9). The tanks shaded by white muslin and unshaded tanks were covered in a sheet of glass, of the same type as that used to build the algae tank, in order to eliminate any possible effects of having the culture tank enclosed. Irradiance was measured using a Skye Intelligent SDL2540 light sensor and ranged from 50 - 400μE m^{-2} sec^{-1} and from 10 to 50μE m^{-2} sec^{-1} in the unshaded and shaded tanks respectively with incident irradiance ranging from 140 - 500μE m^{-2} sec^{-1}.

After a period of three weeks, light response curves for *Littorella* leaves were determined using the protocol outlined in section 2.2a. At the end of the experiment, and 30 days after the application of shading, all above-ground biomass was harvested then prepared for $\Delta^{13}C$ analysis as described in section 2.2d

Section 3
RESULTS

3.1 Rate of Adaptation of *Littorella* to Shading

Total chlorophyll content in both the control and shaded tanks were the same both at the start of the experiment and two days after the application of shading. The shaded tank showed an increase in total chlorophyll levels on days 3, but analysis of variance revealed there to be no significant difference from control total chlorophyll levels until day 9 (Figure 6.4). There is no significant difference between any of the control chlorophyll levels. A summary of the results is shown in Table 6.1.

Both at the start of the experiment and after a period of 12 days there was no significant difference between the chlorophyll *a:b* of shaded and control plants.

3.2 Results of Curve Fitting

The use of all the data points obtained in photosynthetic light curves resulted in a poor fit with a high residual value for points with an oxygen evolution of less than 2 μmol g^{-1} dry weight (DW) s^{-1}. In nearly all cases only the first data point was less than 2 μmol g^{-1}DW s^{-1}, the omission of this data point resulted in a curve that more closely resembled that of the recorded figures and with no data points of high residual value.

Figure 6.5 shows the effect of removing the first data point on the resultant curve. Two of the data sets contained points other than the first point that had high residual values and were rejected. In all cases 95% of the variation was explained by the regression which was highly significant ($P < 0.005$)

The photosynthesis light-response curves used in the analysis are presented in Figure 6.6. Data obtained from these curves are summarised in Table 6.2. One-way analysis of variance was carried out using Genstat 5 and comparisons of data within significant data sets were carried out using orthogonal comparisons (Sokal & Rohlf, 1981).

Maximum gross photosynthesis (Pm) was significantly higher in control *Littorella* plants than in shaded. There was however, no difference in Pm between the types of shading applied (see Table 6.2). Also, there were no significant differences between respiration rates of shaded and unshaded plants. Analysis of the entire data set revealed no significant variation in the reaction constant (Km). However orthogonal contrasts revealed there to be a significant difference in Km between control and shaded *Littorella* plants ($P = 0.05$) and no significant difference between the two shade treatments.

3.3 $\Delta^{13}C$, % Nitrogen and % Carbon

There was no difference in carbon content of shaded and unshaded plants. Nitrogen content increased by 26% 30 days after the application of shading ($P < 0.000$). The $\Delta^{13}C$ values were lower in shaded *Littorella* than in unshaded ($P < 0.000$). Shaded plants had a mean $\Delta^{13}C$ of 29.6‰ and 29.3‰ for *Ulva* and muslin shading respectively and unshaded plants a $\Delta^{13}C$ value of 26.1‰. There was no significant difference between the two shading treatments in terms of nitrogen content, carbon content or $\Delta^{13}C$.

3.4 Chlorophyll Concentration and Chlorophyll *a*:*b* Ratio

One-way analysis of variance of total chlorophyll levels showed a highly significant ($P = 0.001$) effect of shading. Orthogonal comparisons revealed there to be differences between shaded and unshaded *Littorella* chlorophyll levels ($P = 0.002$) and differences between the type of shading ($P = 0.021$): *Littorella* plants that had been shaded by *Ulva* had the highest total chlorophyll concentration.

Chlorophyll *a*:*b* ratios were significantly different between shaded and unshaded plants ($P = 0.027$). There was, however, no difference in chlorophyll *a*:*b* between the type of shading applied ($P = 0.280$).

Figure 6.8 illustrates the differences in total chlorophyll and chlorophyll *a*:*b* ratio on the application of shading. Data are summarised in Table 6.4 and raw data are presented in Appendix F.

Figure 6.4 Shading-induced change in total chlorophyll levels

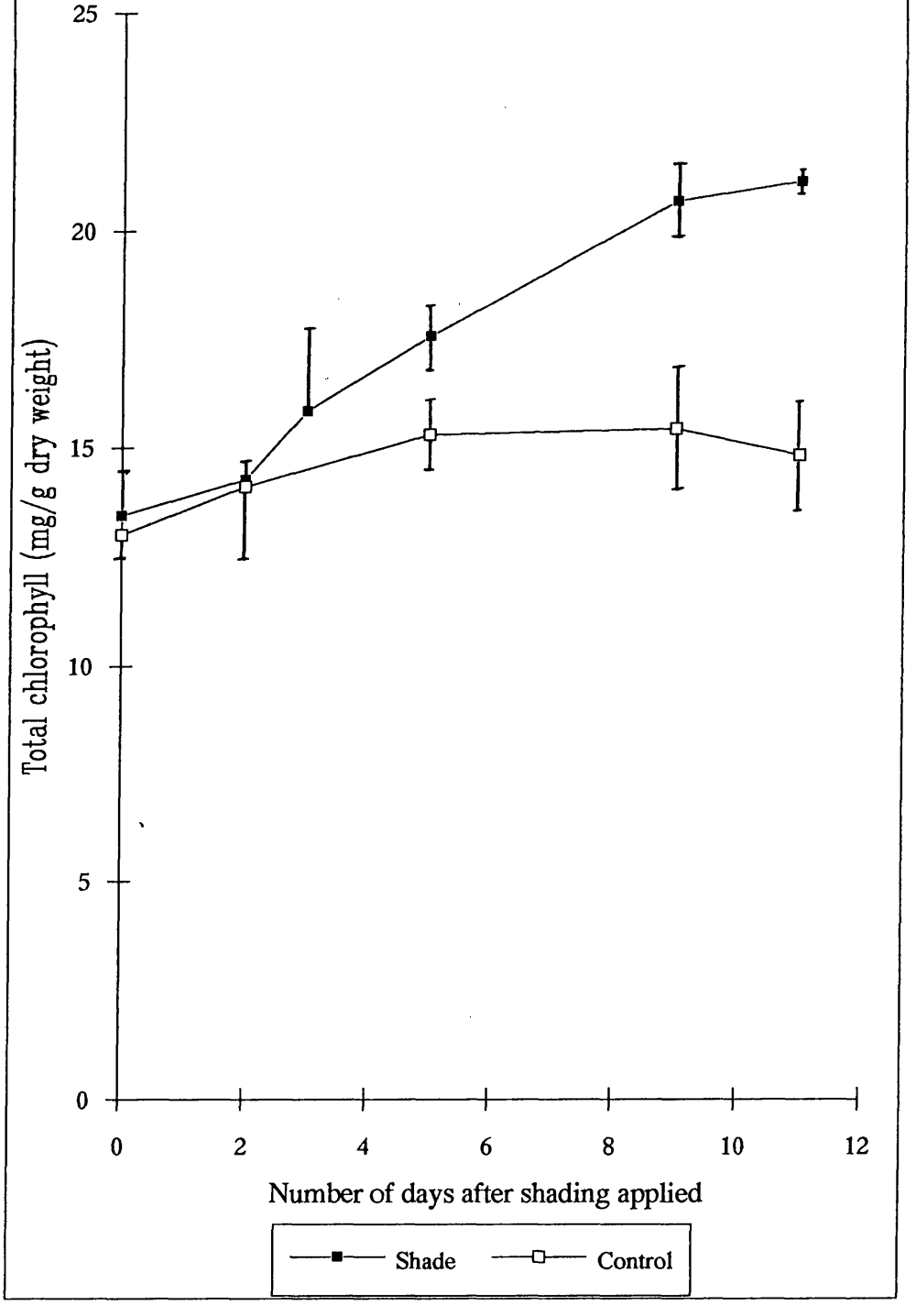


Table 6.1
Chlorophyll content of leaves of *Littorella* on the application of shading.

Treatment	Day no.	Mean total chlorophyll	Standard error	P
Shade	0	13.45	0.41	.698NS
Control	0	13.00	1.02	
Shade	2	14.26	0.30	.934NS
Control	2	14.11	1.70	
Shade	3	15.83	2.23	.563NS
Control	2	14.11	1.70	
Shade	5	17.57	0.73	.077NS
Control	5	15.29	0.70	
Shade	9	20.67	0.88	.033*
Control	9	15.43	1.38	
Shade	11	21.11	0.22	.004**
Control	11	14.82	1.27	

Chlorophyll content expressed as mg g^{-1} dry weight.

NS - not significant

* - $P < 0.05$

** - $P < 0.01$

Figure 6.5 The effect of omitting low oxygen evolution values on shape of resultant curve

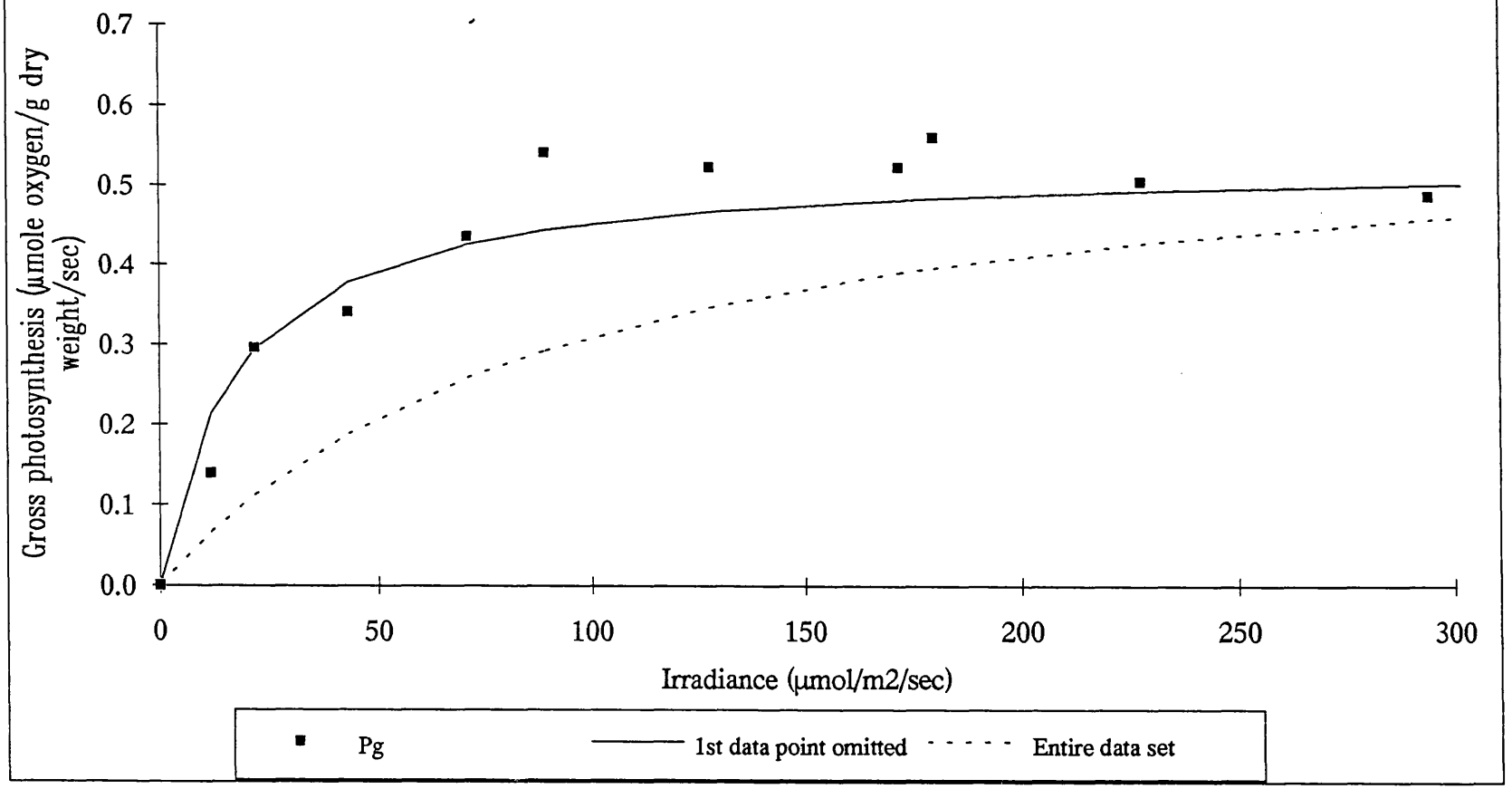
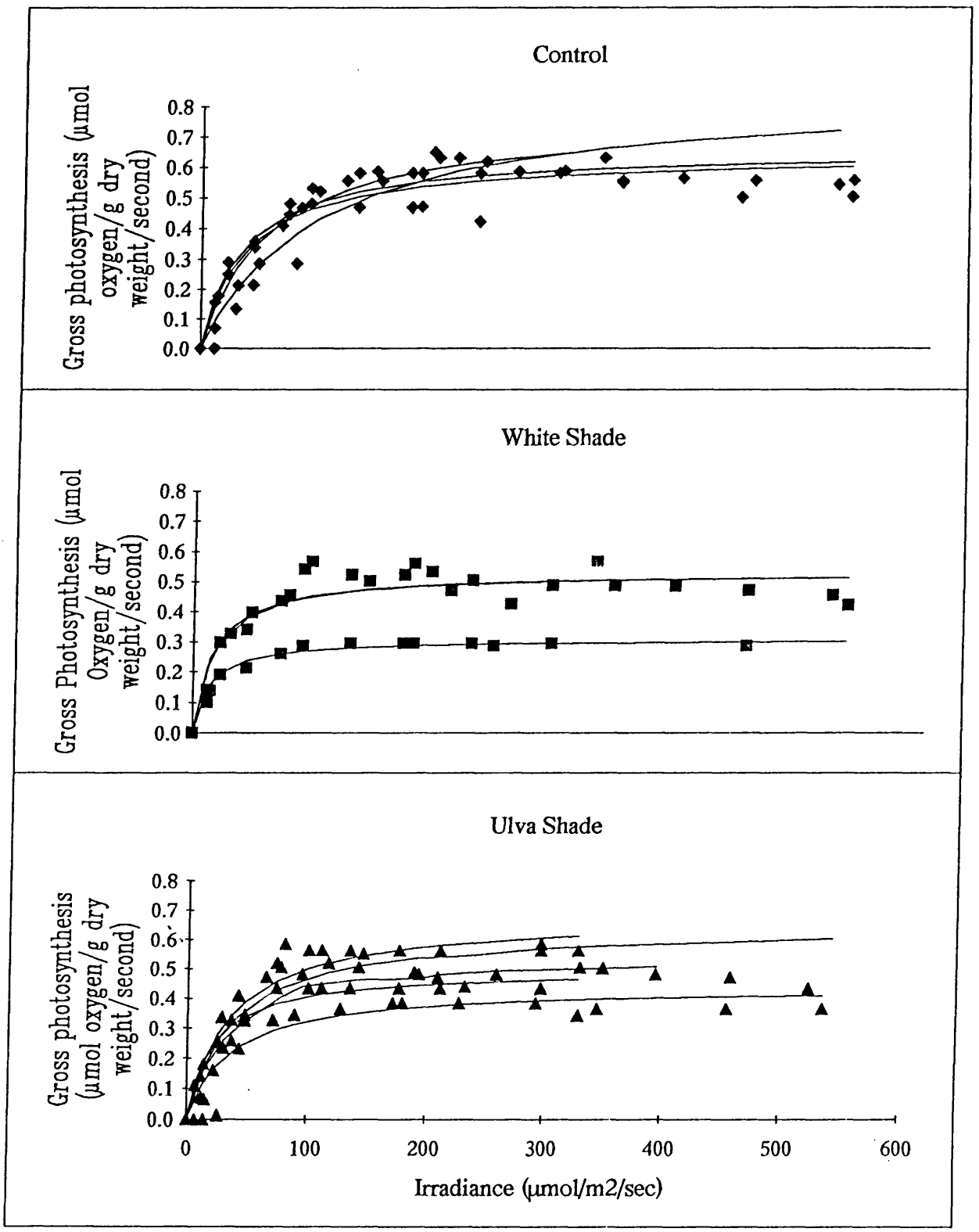


Figure 6.6 Photosynthetic light response curves of Littorella after three weeks shading



Key: Lines derived values
Symbols measured values

Table 6.2a
Summary of photosynthetic characteristics of shaded and unshaded *Littorella* plants

Treatment	Dark Respiration $\mu mol\ O_2\ g^{-1}\ s^{-1}$ mean (s.e.)	Pg max. $\mu mol\ O_2\ g^{-1}\ s^{-1}$ mean (s.e.)	Km $\mu E\ m^{-2}\ s^{-1}$ mean (s.e.)
Control	-0.099(0.040)	0.724(0.049)	54.83(13.33)
Ulva shade	-0.105(0.026)	0.539(0.040)	36.67(7.75)
White shade	-0.118(0.030)	0.456(0.074)	15.73(1.29)

Table 6.2b
Summary of analysis of variance, with orthogonal contrasts, of photosynthetic characteristics

Factor	Comparison	Probability
Respiration	all	0.934^{NS}
Pg max	all	0.017^{*}
	Control vs Shaded	0.007^{**}
	Ulva vs White	0.294^{NS}
Km	all	0.082^{NS}
	Control vs Shaded	0.050^{*}
	Ulva vs White	0.233^{NS}

^{NS} - not significant
* - significant ($P < 0.05$)
** - significant ($P < 0.01$)

Table 6.3a Summary of $\delta^{13}C$, % nitrogen and % carbon levels in shaded and unshaded leaves of *Littorella*

TREATMENT	%N	se	%C	se	δ PDB	se
Ulva	2.71	.038	38.82	.97	-29.58	.15
Muslin	2.68	.028	40.43	.33	-29.28	.30
Unshaded	2.02	.072	39.08	.65	-26.11	.46

Table 6.3b Summary of analysis of variance of ANCA-MS results

% nitrogen TREATMENT	P
Ulva vs Muslin	0.456^{NS}
Ulva vs Control	0.000****
Muslin vs Control	0.000****
Ulva vs Muslin vs Control	0.000****
% carbon	
TREATMENT	
Ulva vs Muslin	0.145^{NS}
Ulva vs Control	0.828^{NS}
muslin vs Control	0.092^{NS}
Ulva vs Muslin vs Control	0.246^{NS}
PBD	
TREATMENT	
Ulva vs Muslin	0.406^{NS}
Ulva vs Control	0.000****
muslin vs Control	0.000****
Ulva vs Muslin vs Control	0.000****

NS - not significant
**** - highly significant ($P < 0.000$)

Figure 6.7 % nitrogen, % carbon and delta-PBD concentration in Littorella leaves three weeks after the application of shading.

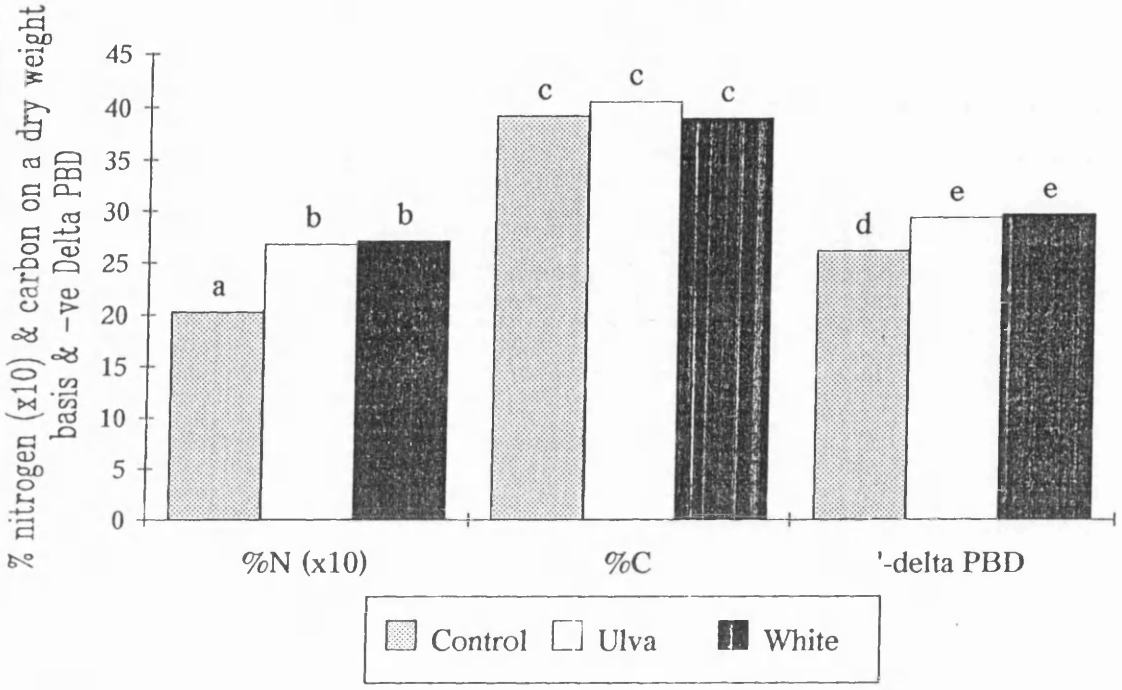
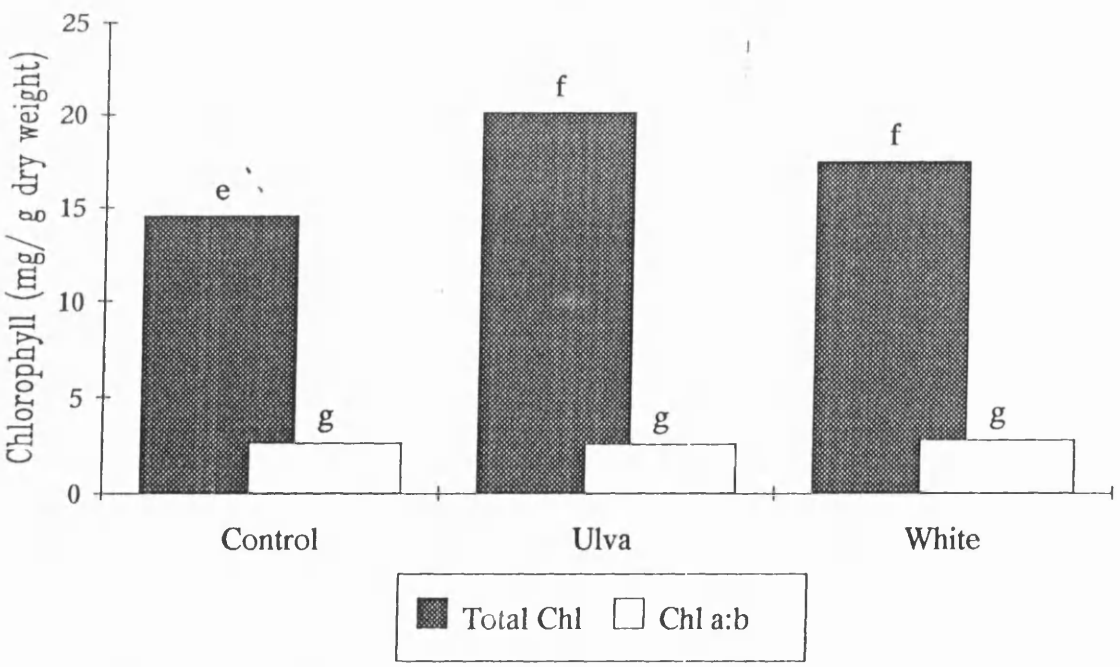


Figure 6.8 Chlorophyll concentration and chlorophyll a:b in shaded and unshaded Littorella



atching letters denote no significant difference at 95%

Table 6.4a
Summary of chlorophyll concentration and chlorophyll *a:b* ratio of leaves after three weeks of shading

Treatment	Total Chlorophyll (s.e.) (mg g^{-1} dry weight)	Chlorophyll *a:b*
Control	14.51 (0.56)	2.62
Ulva shade	20.09 (1.03)	2.53
White shade	17.44 (0.99)	2.75

Table 6.4b
Summary of analysis of variance, with orthogonal comparisons, of chlorophyll concentration and chlorophyll *a:b* three weeks after the application of shading

Factor	Comparison	Probability
Total Chlorophyll	all	0.001^{***}
	Control vs Shaded	0.002^{**}
	Ulva vs White	0.021^{*}
Chlorophyll *a:b*	all	0.051^{NS}
	Control vs Shaded	0.027^{*}
	Ulva vs White	0.280^{NS}

^{NS} - not significant
* - significant ($P<0.05$)
** - highly significant ($P<0.01$)
*** - very highly significant ($P<0.001$)

Section 4
DISCUSSION

4.1 Change in Chlorophyll Concentration

Although there was a change in total chlorophyll concentration 9 days after the application of shading, there was neither a decrease in the chlorophyll *a:b* after 12 days, nor a change in the chlorophyll *a:b* in the second experiment where shading was applied for 3 weeks. A decrease in chlorophyll *a:b* was observed in plants experiencing shading for 6 weeks (Chapter 5), and in field populations growing under filamentous algal mats (Chapter 4).

Søndergaard & Bonde (1988) observed an increase in the total chlorophyll content of leaves of shallow water (0.2m) *Littorella* plants between the months of August and November, but no changes in chlorophyll *a:b* occurred. There were however significant differences in the chlorophyll *a:b* deep water (2.3m) and shallow plants. The chlorophyll *a:b* data presented in Chapter 6 are similar to those measured in the deep-water populations of *Littorella* in the above paper.

The data presented in this thesis suggest that a decrease in chlorophyll *a:b* is a slower response to shading in *Littorella* than an increase in both chlorophylls *a* and *b*. In order to confirm this, an experiment is required, where the chlorophyll content of shaded and unshaded *Littorella* plants is measured regularly for up to six weeks after the application of shading. However, Robe & Griffiths (1990) found no differences in chlorophyll *a:b* of *Littorella* plants grown at 50 and 300 $\mu E\ m^{-2}\ s^{-1}$ for between 5 and 11 months.

An increased chlorophyll concentration will result in a greater proportion of incident light being absorbed by the leaf (Björkman, 1981). Pizarro & Montecino (1992) demonstrated that field populations of *Elodea potamogeton* maintained constant photosynthesis and growth rates throughout the year by altering chlorophyll *a* concentration in the leaves. Other than light regime, the study plants experienced constant physical and chemical conditions. Change in chlorophyll *a* concentration was apparent within a few days, and there was an inverse relationship

between chlorophyll *a* concentration and the total solar irradiation available on the three days prior to sampling.

4.2 Effects of Shading Material

In all the measurements carried out the only significant difference between the muslin shading and *Ulva* shading was in total chlorophyll levels. Data presented earlier in this chapter suggested that change in total chlorophyll concentration was a more rapid response to shading than changes in chlorophyll *a:b*. It is possible that any effects due to alteration of the proportion of incident far red light may not yet be apparent. However, from the data presented in this chapter there is no evidence against the use of neutral density materials in measurements of changes in photosynthetic response to PAR after the application of shading.

4.3 Interpretation of Photosynthetic Light Curves.

It could be expected, that the measurement of oxygen evolution of submerged macrophytes in air, as opposed to under water, would overestimate photosynthesis as carbon dioxide will not be limiting due to boundary layer effects. Values for the maximum gross photosynthetic rate are similar to those measured by Nielsen & Sand-Jensen (1989): values of 7.3×10^{-8} and 4.7 to 7.2 x 10^{-8} moles O_2 $g^{-1}DW$ s^{-1} for Nielsen & Sand-Jensen and this study respectively. Contrary to Westlake (1967), Robe & Griffiths (1988) demonstrated that *Littorella* photosynthesis was not carbon limited *in-vitro*, and that uptake of carbon dioxide was primarily through the roots (Sand-Jensen & Søndergaard, 1978; Robe & Griffiths, 1990). Uptake of carbon dioxide is carried out by chloroplasts lining the gas-filled lacunae (Raven, 1970), consequently an increase in photosynthetic rate due to reduced the diffusion resistance of carbon dioxide in air, as opposed to water, (Smith & Walker, 1980) did not occur.

The lack of difference between dark respiration rates is contrary to what would be expected of a shade-adapted plant. Shade-adapted plants tend to have lower dark respiration rates and the compensation point between respiration and photosynthesis is consequently lowered (Björkman, 1981).

The lower Michaelis-Menten constant (Km) for photosynthesis in shaded *Littorella* plants results in more efficient photosynthesis at lower light levels. Low PAR adapted *Littorella* had a lower maximum photosynthetic rate (Pm) than the unshaded plants, so were not so efficient at higher irradiances than unshaded plants.

The increase in nitrogen content of leaves reflects an increase in photosynthetic apparatus as described in Chapter 4 after filamentous algal mat formation.

In this experiment, photosynthesis measurements were carried out the leaves orientated perpendicular to the light source: an occurrence not encountered under natural conditions. Light intensity experienced by a normally positioned leaf can be calculated by multiplying the measured light by the cosine of the angle of incidence (Duncan *et al.*, 1967). The measurement of leaf orientation in response to shading was not carried out in this work. Further work in this area may prove interesting.

4.4 $\Delta^{13}C$ Measurements

In aquatic macrophytes, $\Delta^{13}C$ values range from -11‰ to -39‰ (Farquhar *et al.*, 1989). Several studies have shown $\Delta^{13}C$ measurements not to be useful in the determination of photosynthetic pathways in aquatic macrophytes (Osmond *et al.*, 1981; Keeley *et al.*, 1986; Keeley, 1988). In each of these studies, plants of different life forms were compared from the same water body.

In comparison to terrestrial systems, the factors governing $\Delta^{13}C$ in freshwaters are complex. As well as isotope discrimination by the enzymes of carbon fixation, differences in diffusion resistance and bicarbonate will also alter $\Delta^{13}C$ (Smith & Walker, 1980). Raven *et al.* (1987) demonstrated that it was possible to distinguish between CAM and non-CAM photosynthesis in aquatic macrophytes of the same morphology, but not between species with different life forms.

Values for $\Delta^{13}C$ obtained in this work are similar to those presented by several authors and summarised in Raven *et al.* (1987) for *Lobelia dortmanna.* There is no

evidence of CAM in *L. dortmanna*, however, even in terrestrial systems there is a large overlap in the $\Delta^{13}C$ values obtained for different species of plant that undergo C3 and CAM photosynthesis (Farquhar *et al.*, 1989).

In aquatic systems the $\Delta^{13}C$ values of plants will reflect that of the surrounding media which is variable (Raven *et al.*, 1987). In *Littorella* carbon uptake is primarily through the roots (Sand-Jensen & Søndergaard, 1978), so tissue $\Delta^{13}C$ values are likely to reflect that of the sediment interstitial water. The measurement of $\Delta^{13}C$ in sediment interstitial water is difficult and time consuming. In a system such as this, where all sediment is from the same source, $\Delta^{13}C$ values are likely to be the same in each tray (A. Johnston, University of Dundee *pers. comm.*). It is therefore possible to make comparative measures of $\Delta^{13}C$ without measuring interstitial sediment $\Delta^{13}C$ levels.

Plants carrying out CAM photosynthesis will have a higher $\Delta^{13}C$ than those utilising the C3 pathway alone (Farquhar *et al.*, 1989). The lower $\Delta^{13}C$ in shaded *Littorella* can be attributed to a reduction in CAM. Madsen (1987b) studied the effects of carbon dioxide, inorganic nutrients and irradiance on CAM by measuring changes in diurnal titratable acidity. CAM was found to be plastic and dependant on the environmental conditions. High levels of free carbon dioxide, or low levels of nutrients resulted in a decrease in CAM. Low levels of free carbon dioxide with high nutrients and irradiance maintained CAM. Under conditions of low irradiance CAM was suppressed even when free carbon levels were low. Low light levels induced a significant decrease in CAM in 3 weeks. Using the same method, Robe & Griffiths (1990), observed higher CAM in *Littorella* plants cultured under higher irradiances than those in more shaded conditions.

The maintenance of CAM is energy-expensive (Nobel, 1991). Madsen (1987b) suggested a reduction of CAM with low irradiance was a good strategy as it allowed optimal allocation of energy to growth, which may be of importance in turbid conditions.

Section 5
CONCLUSION

Littorella showed a significant increase in total chlorophyll, 9 days after the application of shading. Six weeks after the application of shading in these greenhouse trials, chlorophyll *a:b* was lower in shade plants when compared with unshaded controls. There were no significant differences between chlorophyll *a:b* of shaded and unshaded *Littorella* plants in the experiment where shading was only applied for 3 weeks. An increase in total leaf chlorophyll concentration was a more rapid response to shading than a decrease in chlorophyll *a:b*.

In comparison with unshaded controls, the photosynthesis light response curves of shade adapted *Littorella* plants had a higher photosynthetic efficiency at low irradiance and a decreased maximum photosynthetic rate. There were no differences in the dark respiration rate of shaded and unshaded *Littorella* plants. Higher $\Delta^{13}C$ levels in shade adapted *Littorella*, in comparison to unshaded controls, indicated a reduction in CAM.

There were no differences in dark respiration rate, photosynthetic efficiency, maximum photosynthetic rate, nitrogen content and $\Delta^{13}C$ of *Littorella* leaves shaded by white muslin, when compared to those shaded with *Ulva*. Thus, there is no evidence to reject the use of neutral density shading in such studies of shading response.

Chapter 7:

Algal Effects on *Littorella uniflora* (L.) Ascherson: Model and Conclusions

Chapter 7:

Algal Effects on *Littorella uniflora* (L) Ascherson: Model and Conclusions

Section 1

EVIDENCE FOR A MODEL OF POPULATION MAINTENANCE IN SCOTTISH LOCHS

The most important abiotic factors affecting *Littorella* distribution in the four study lochs were exposure and sediment organic content (Chapter 4). Although *Littorella* is well adapted to exposure (Keddy, 1983), in the four lochs in this study, *Littorella* plants from sites with low exposure ratings were larger in terms of dry weight and were also more abundant than *Littorella* plants from more exposed sites. *Littorella* plants from sites with a higher organic content had a greater biomass than those from those with a lower organic content (Chapter 4).

Chapter 4 presents evidence that the most important algal component affecting *Littorella* performance in the four target lochs was filamentous algae. In the absence of filamentous algal biomass data, phytoplankton chlorophyll *a* levels appeared to have an effect. However in Lake of Menteith the effect of phytoplankton alone was not sufficient to bring about any change in the chlorophyll content in the leaves of *Littorella*. Given the lack of morphological plasticity in the leaves of *Littorella* (Søndergaard & Bonde, 1988) change in leaf chlorophyll concentration is the most obvious indicator of shade adaptation in *Littorella*.

Chapter 5 demonstrated that *Littorella* can withstand periods of severe shading (at least those experienced under a filamentous algal bloom - e.g. Robe & Griffiths, 1992) with no loss in biomass, by physiological adaptation to low light levels. These include a reduction in CAM; increased leaf chlorophyll and nitrogen levels; decreased chlorophyll *a:b*; higher photosynthetic efficiency at low irradiance and a decreased maximum photosynthetic rate.

The negative regression between leaf number and filamentous algal biomass,

suggests that in the field, some leaf loss may occur in the presence of filamentous algae (Chapter 4). After the removal of shading, *Littorella* can undergo relatively rapid growth and after a period of a few weeks establish a biomass this is no different from plants that have not experienced shading. The leaves of these unshaded plants however are shorter and more numerous than those of plants that have not experienced deep shading in their life history.

Filamentous algal mats are not present throughout the year. In this study filamentous algae in Loch of Lowes and Loch Dee were present from June in both 1990 and 1991, and were absent by November 1990. In the study by Robe & Griffiths (1992) filamentous algal mats were present during the months of May and June only.

Several studies have concluded that light is the most important factor in aquatic macrophyte productivity (Boylen & Sheldon, 1976; Wium-Andersen & Borum, 1984; Farmer & Spence, 1987). In the northern hemisphere maximal daily insolation occurs during the month of June (Kirk, 1983) during which time the algal mats in Lochs Dee and Lowes are developing. Evidence from other studies suggest that the presence of filamentous algae from June to November will reduce irradiance at periods when, in the absence of filamentous algae, *Littorella* growth rates are maximal. For example, Farmer & Spence (1987) found the growth rate of *Lobelia dortmanna* to increase significantly in late spring, with maximal leaf production occurring in July. Sand-Jensen & Søndergaard (1978), in leaf marking experiments, found that maximum leaf turnover rate in *Littorella* occurred in June-July, in a lake that did not experience algal loading; however a net increase in biomass occurred throughout the year. There are several studies on a range of macrophyte species that provide evidence of net productivity in winter (e.g. Boylen & Sheldon, 1976; Sand-Jensen & Søndergaard, 1978; Farmer & Spence, 1987). In this study, leaf marking experiments in the field were not successful, as the blanketing effect of filamentous algae was under-estimated, and 'day-glo' orange ribbon was not a sufficient marker to allow the relocation of experimental pots.

As no plants actually died in any of the shading treatments, there is no evidence

from this study to suggest that the presence of filamentous algae would result in the loss of *Littorella* from Scottish lochs. It is hypothesised that although the presence of filamentous algal mats results in a great reduction in irradiance, *Littorella* can withstand low light levels due to physiological adaptation. Net photosynthesis at other times of the year in the absence of filamentous algae is sufficient to maintain the population.

Section 2
THE PROPOSED MODEL

Figure 7.1 presents a model for the maintenance of populations of *Littorella* in the lochs studied based on the data described in Chapters 2 to 6, and on information from the available literature on *Littorella.*

In the absence of algal loading, *Littorella* is primarily affected by the sediment organic content and exposure rating, with more densely populated stands of *Littorella* occurring in more sheltered sites with a high organic content. In Chapter 5 data were presented to demonstrate that *Littorella* showed a non-linear response to an incease in sediment organic content. It is, therefore, likely that in conditions when sediment organic matter increases to concentrations greater than that in the four sites in this study, this relationship may break down, possibly due to competition with macrophytes better adapted to higher nutrient conditions.

The presence of phytoplankton may result in some physiological adaptation in response to shading; however this is slight in comparison to the effects of filamentous algae. The onset of algal shading in June will result in a physiological (and possible morphological) adaptation that will allow *Littorella* to survive the period of low irradiance with little or no loss in biomass. After the die-back of filamentous algae in the autumn the increased irradiance may allow net production to occur (e.g. Sand-Jensen & Søndergaard, 1978) and the increasing irradiance in the spring will provide sufficient light for rapid growth of many shorter leaves in comparison to unshaded populations. The population is consequently maintained by winter and spring growth.

Loss of macrophytes may occur in the event of either of two occurrences:

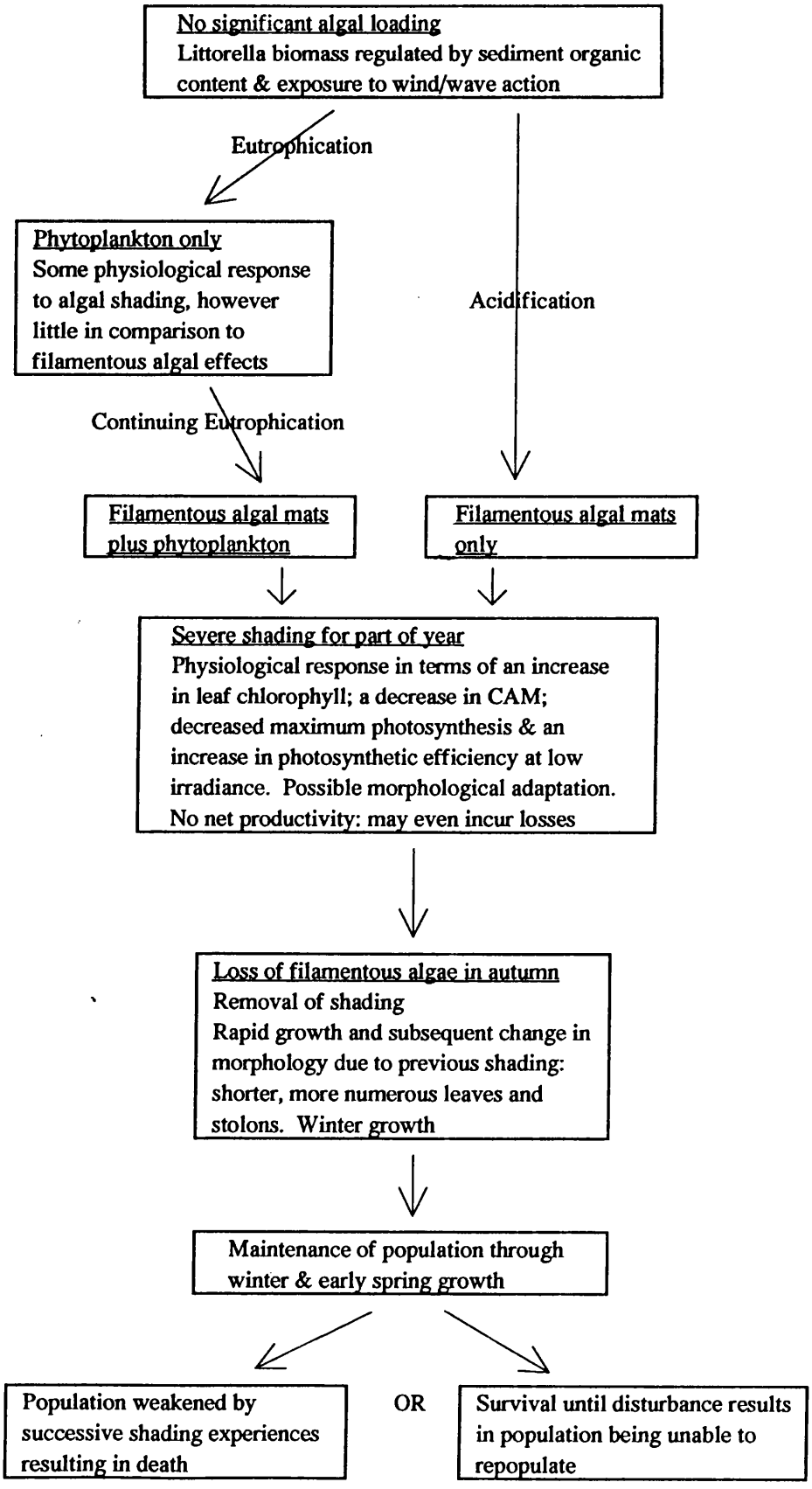
1. Due to the presence of filamentous algal mats, populations of *Littorella* may become gradually weakened by successive years with long periods of low irradiance. This will result in poor growth rates and low recolonisation rates until the population eventually fails to maintain itself.

Titus & Hoover (1993) exposed two freshwater macrophytes (*Najas flexilis* and *Vallisneria americana*) to low pH. Both species showed reduced population maintenance. *N. flexilis* had reduced number of seeds produced per plant and *V. americana* produced fewer smaller tubers. These authors proposed the closing spiral hypothesis whereby growth at low pH resulted in progressively fewer seeds or reduced stolon production and with each summer the plants would become smaller, until one year the plants would be unable to maintain the population. Their findings were backed up by field transplant experiments.

2. The population may survive and be able to maintain itself; however, in the event of a catastrophic destruction of the population as a whole or in part, recolonisation will not be possible.

Scheffer (1990) presented simple models in which lakes are predicted to exist in a series of stable states, and that the transition between for example clear water and turbid water due to algal growth will be rapid in shallow lakes. Algal growth will increase with nutrient enrichment; however the presence of aquatic macrophytes has a negative effect on turbidity. Macrophytes will not be affected by the increase in algal growth, until algal biomass levels are high and then the macrophytes will disappear suddenly, resulting in increased algal growth.

Figure 7.1 Model for the maintenance of Littorella populations in Scottish lochs



Section 3

FUTURE WORK: TESTING THE MODEL

The work described here presents evidence that *Littorella* populations can be maintained under conditions of severe algal loading. Further questions need to be addressed in order to refine the model.

1. Does *Littorella* achieve net production under a filamentous algal mat? Greenhouse experimental data suggests that at light levels of 10-40 μE m^{-2} s^{-1}, *Littorella* will undergo no net production over a period of six weeks. The form of shading in a greenhouse experiment is continuous, whereas filamentous algae may be moved by wave action and water currents resulting in small bursts of light. It is possible that *Littorella* may be able to utilise rapidly such bursts of light in a manner rather similar to the way in which forest understorey plants can respond rapidly to sunflecks (Pearcy, 1990). *In situ* growth experiments of *Littorella* grown under algal mats for several months, such as those in Loch Dee and Loch of Lowes are required.

Ozimek *et al.*, (1991) found that both *Potamogeton pectinatus* and *Elodea canadensis* showed significantly reduced growth, and an increased rate of decomposition when grown with the filamentous algae *Cladophora glomerata* under laboratory conditions. In their discussion, these authors presented data that illustrates the variability of filamentous algal dispersal in natural systems (400% differences in shading in 3 minutes) and suggest that this variability may reduce the effects of filamentous algae on aquatic macrophytes.

2. If *Littorella* does not undergo a net increase in biomass under the cover of an algal mat, field rates of leaf production in the winter would be required in order to determine if winter and spring production rates are sufficient to maintain and/or expand the field population.

3. It would be interesting to test whether under conditions of successive shade and exposure to light the population does become weakened. In this case greenhouse

experimentation comparing unshaded populations of *Littorella* with populations exposed to alternating successive shade/light regimes would be required.

4. Nobel *et al.* (1993), suggested that plant canopy structure was of greater importance to productivity than adaptation of the photosynthesis to different irradiance levels. Due to lack of structural support, canopy structure measurements may be difficult in the majority of aquatic macrophyte species. In isoetids, which have stiff leaves that retain their orientation once the support of the surrounding water has been removed, measurement of leave orientation is possible. Comparisons of the orientation of shade and non-shade populations of *Littorella* may provide some interesting results.

Section 4
CONCLUSIONS

The main findings of this study are as follows:

Chapter 1

Under conditions of both acidification and eutrophication the relative abundances of macrophytes and algae in lentic freshwater systems will alter. In both cases an increase in filamentous algal mat formation may occur. Changes in epiphyte abundance and phytoplankton density will also occur. A decrease in macrophyte species diversity and abundance may occur, with the eventual loss of macrophytes from systems experiencing either eutrophication or acidification.

Chapters 2 and 3

Detrended Correspondence Analysis demonstrated that the four lochs in the study were representative of one community type. A total of 15 macrophyte species were recorded along the 1m isobath. The four most commonly occurring species were: *Isoetes lacustris*; *Lobelia dortmanna*; *Littorella uniflora* and *Myriophyllum alterniflorum*. The four lochs studied experienced a range of algal loadings. Loch Dee was acidic and experienced algal mat formation. Loch Lomond was oligotrophic and had very low algal loading. Lake of Menteith was mesotrophic/eutrophic and experienced phytoplankton blooms in the spring and autumn. Loch of Lowes was fairly eutrophic with phytoplankton chlorophyll *a* in excess of 10 $\mu g\ l^{-1}$ and extensive filamentous algal mats were formed from June until November.

Chapter 4

Of the three algal components considered in this study, stepwise linear multiple regression revealed filamentous algal biomass to explain the most variation in *Littorella* field attributes. *Littorella* plants growing under filamentous algal mats tended to have a higher total leaf nitrogen and chlorophyll content; a lower chlorophyll *a:b*; a greater number of stolons per plant and fewer leaves per plant. Epiphytes did not have a significant effect on the measured attributes of *Littorella*

in the four lochs in this study.

In this chapter, phytoplankton chlorophyll *a* only became important in explaining the variation in *Littorella* field attributes when data concerning filamentous algal biomass was not available. It was concluded in studies such as this, if filamentous algae were present, but not included in the data set, the influence of phytoplankton on the macrophyte flora may be over-estimated.

The most important abiotic factors related to variation in *Littorella* attributes were sediment organic content and exposure rating. More exposed sites tended to have fewer smaller *Littorella* plants. Sites with a high sediment organic content tended to have a greater biomass of all species of aquatic macrophyte and *Littorella.*

The percentage of the variation of *Littorella* morphological attributes explained by the measured environmental variables was low: this was attributed to a lack of morphological plasticity of isoetids which has been reported by several authors (e.g. Holstrup & Wiegleb, 1991b; Wilson, 1991). In contrast, more than 65% of the measured variation in physiological adaptations, such as changes in leaf chlorophyll and leaf nitrogen, could be attributed to filamentous algal biomass.

Chapter 5

Littorella demonstrated a non-linear growth response to increased sediment organic matter content, with maximum growth occurring at intermediate concentrations of sediment organic matter.

After 6 weeks of shading *Littorella* plants did not increase in biomass, but did show some morphological adaptation: shaded plants had a higher Specific Leaf Area than unshaded controls. Shade plants also had a higher leaf chlorophyll content and a lower chlorophyll *a:b*. After the removal of shading *Littorella* plants that had been shaded produced a greater number of shorter leaves in comparison to unshaded controls.

Chapter 6

In this chapter physiological responses of *Littorella* to shading were considered. Total leaf chlorophyll showed a significant increase 9 days after the application of shading in comparison to unshaded controls. In a second experiment in this chapter chlorophyll extraction were carried out 3 weeks after the application of shading. No change in chlorophyll *a:b* was observed in either of these experiments. A decrease in chlorophyll *a:b* had been attributed to filamentous algal mat formation in Chapter 4. In chapter 5 a lower chlorophyll *a:b* ratio had been observed in shaded plants in comparison to unshaded controls 6 weeks after the application of shading. It was concluded that a decrease in chlorophyll *a:b* was a slower response to shading than an increase in total chlorophyll.

Measurements of photosynthesis light response curves using a gas phase oxygen electrode showed that shade-adapted *Littorella* plants had a higher photosynthetic efficiency at low irradiances and a lower maximum rate of photosynthesis in comparison to unshaded controls. There were, however, no differences in dark respiration rates. Shade adapted *Littorella* plants also showed a decrease in CAM as indicated by a higher $\Delta^{13}C$.

A comparison of live algae and muslin as shading materials revealed no significant difference in the above photosynthetic characteristics of *Littorella.* It was concluded that there was no evidence to reject the use of neutral density shading materials in such studies of shading response.

Chapter 7

In this chapter a model for the maintenance of *Littorella* populations in Scottish lochs was presented. This model was based on the results of Chapters 2-to-6 and available information in the published literature.

In the model filamentous algae were considered to be the most important algal component affecting *Littorella* in Scottish lochs. In different systems, with different species that have different physiology to *Littorella,* it is likely that the other algal components considered in this study may assume greater importance.

Summary of site algal problems

Lomond

None

Dee

Filamentous algae

Menteith

Phytoplankton

Cladophora balls

Lowes

Phytoplankton

Filamentous algae

Bibliography:

BIBLIOGRAPHY

ADAMS, M.S., TITUS, J.E., McCRACKEN, M.D. (1974): Depth distribution and photosynthetic activity in a *Myriophyllum spicatum* community in Lake Wingra. Limnology & Oceanography, 19:377-389.

ALEYA, L., DEVAUX, J., EL MAGOURI, H., MARVALIN, O., AMBLARD, C. (1988): The usefulness of several methods for the estimation of phytoplankton biomass. European Journal of Protistology, 23(4):334-342.

ALLEN, J.R. (1985): Principles of Physical Sedimentology. George, Allen & Unwin. 272pp.

ALLEN, S.E., GRIMSHAW, H.M., ROWLAND, A.P. (1986): Chemical Analysis. Chapter 6 in: Methods in Plant Ecology. Eds: P.D. Moore, S.B. Chapman. Blackwell Scientific Publications pp285-344.

ALLENBY, K.G. (1981): Some analyses of aquatic plants and their waters. Hydrobiologia, 77:177-189.

ARTS, G.H.P., DE HAAN, A.J., SIEBURN, M.B., VERGHEGGEN, G.M. (1989): Extent and historical development of the decline of Dutch soft waters. Proceedings of the Koninklijk Nederlandse Akademide van Wetenschappen Series C, Biological and Medical Sciences 92(3):281-298.

ARTS, G.H.P. & VAN DER HEIJDEN, A.J.M. (1990): Germination Ecology of *Littorella uniflora* (L.) Aschers. Aquatic Botany, 37:139-151.

ARTS, G.H.P., ROELOFS, J.G.M., DE LYON, M.J.H. (1990a): Differential tolerances among soft watered macrophyte species to acidification. Canadian Journal of Botany, 68:2127-2134.

ARTS, G.H.P., VAN DER VELDE, G., ROELOFS, J.G.M., VAN SWAAY, C.A.M. (1990b): Successional changes in the soft-water macrophyte vegetation of (sub) atlantic, sandy, lowland regions during this century. Freshwater Biology, 24:287-294.

AULIO, K. (1985): Differential expression of diel metabolism in two life forms of *Littorella uniflora* (L.) Aschers. New Phytologist, 100:533-536.

AULIO,, K. (1986): CAM-like photosynthesis of *Littorella uniflora* (L.) Aschers. Annals of Botany, 58:273-275.

AZCON-BIETO, J. (1992): Relationships between photosynthesis and respiration in the dark in plants. Chapter 19 In: Trends in Photosynthesis Research. Ed: J. Barber, M.G., Guerrero, H. Medrano. Pub: Intercept Andover, Hampshire. Pp 241-254.

BAILEY-WATTS, A.E., KIRIKA, A. (1991): A Basic Surveillance of the Phytoplankton of a Rapidly Flushed Upland Lake (Loch Dee, Galloway, Scotland), and Thoughts on the Ecological Determinants of the Main Features of the Algal Assemblages. Unpublished report to the Scottish Office, Department of Agriculture and Fisheries.

BAILEY-WATTS, A.E., KIRIKA, A. (1993): Phytoplankton and controlling factors in the rapidly flushed, upland Loch Dee (Galloway, Scotland). In: Proceedings of the Loch Dee Symposium. Acidification, Forestry & Fisheries Management in Upland Galloway. Foundation for Water Research. Pp 83-96.

BALLS, H., MOSS., B., IRVINE, K. (1989): The loss of submerged plants with eutrophication I. Experimental design, water chemistry, aquatic plant and phytoplankton biomass experiments carried out in ponds. Freshwater Biology, 22:71-87.

BARKO, J.W., FILBIN, G.J. (1983): Influences of light and temperature on chlorophyll in submerged freshwater macrophytes. Aquatic Botany,15:249-255.

BARKO, J.W., GODSHALK, G.L., VIRGINIA, C., RYBICKI, N.B. (1988): Effects of submersed aquatic macrophytes on physical and chemical properties of surrounding water. US Army Engineers Waterways Experimental Station, Vicksburg. US. Technical Report A-88-11.

BATTARBEE, R.W. (1984): Diatom analysis and the acidification of lakes. Philosophical Transactions of the Royal Society of Edinburgh B, 305:451-477.

BATTARBEE, R., FLOWER, R., STEVENSON, A.C., RIPPEY, (1985): Acidification of Lochs. Nature, 314:350-352.

BATTARBEE, R.W., LOGAN, N.A., MURPHY, K.J., RAVEN, P., ASTON, R.J., FOSTER, G.N. (1991): Other Aquatic Biology. In: Restoring Acid Waters: Loch Fleet 1984 - 1990. Ed: G. Howells, T.R.K. Dalziel. Elsevier Applied Science, London. Pp 389-330.

BEST, E.P., DE VRIES, D., REINS, A. (1984): The macrophytes in the Loosdrecht Lakes: a story of their decline in the course of eutrophication. Verh. Internat. Verein. Limnol., 22:868-875.

BILLINGTON, N. (1991): A comparison of three methods of measuring phytoplankton biomass on a daily and season basis. Hydrobiologia, 226:1-15.

BJÖRKMAN, O. (1981): Responses to different quantum flux densities. In: Encyclopedia of Plant Physiology New Series. Volume 12a. Physiological Plant Ecology I. Responses to the Physical Environment Ed: O.L. Lange, P.S. Nobel, C.B. Osmond, H. Zielger. Springer-Verlag. Pp 57-107.

BLACKMORE, S., TOOTHILL, E. (editors) (1984): Penguin Dictionary of Botany. Penguin Great Britain, 391pp.

BLANQUIST, S. (1990): Sampling performance of Ekman grabs - in situ observations and design improvements. Hydrobiologia, 190:245-254.

BODKIN, P.C., POLUSZNY, U., DALE, M. (1980): Light and pressure in two freshwater lakes and their influence on the growth, morphology and depth limits of *Hippuris vulgaris*. Freshwater Biology, 10:545-552.

BOSTON, H.L. & ADAMS, M.S. (1983) Evidence of crassulacean acid metabolism in two north American isoetids. Aquatic Botany, 15:381-386.

BOSTON, H.L. & ADAMS, M.S. (1986): The contribution of crassulacean acid metabolism to the annual productivity of two aquatic vascular plants. Oecologia, 68:615-622.

BOSTON, H.L. (1986): A discussion of the adaptations for carbon acquisition in relation to growth strategy of aquatic isoetids. Aquatic Botany, 26:259-270.

BOSTON, H.L., ADAMS, M.S., MADSEN, J.D. (1989): Photosynthetic strategies and productivity in aquatic systems. Aquatic Botany, 34:27-57.

BOYLEN, R.B., SHELDON, C.W. (1976): Submergent macrophtes: growth under winter ice cover. Science, 194:34-38.

BOWES, G. (1987): Aquatic plant photosynthesis: strategies that enhance carbon gain. In: Plant Life in Aquatic and Amphibious Habitat, special Publication No.5. of British Ecological Society. Ed: R.M.M. Crawford. Blackwell Scientific Publications. 452pp.

BOWN, C.J., SHIPLEY, B.M., BIBBY, J.S. (1982): Soil Survey of Scotland Sheet 6. Soil and Land Capability for Agriculture, south-west Scotland. Macaulay Institute for soil research. Aberdeen University Press.

BREMNER, J.M.M BREITENBECK, G.A. (1983): A simple method for determination of ammonium in semi-micro Kjeldal analysis of soil and plant material using a block digestor. Communications in Soil Science and Plant Analysis, 14:905-913.

BREMNER, J.M., MULVANEY, C.S. (1982): Nitrogen-total. In: Methods of Soil Analysis Part 2. Chemical & Microbiological Properties. Agronomy Monograph no. 9 ASA-SSA, Madison.

BROCK, T.D., (1983): Membrane Filtration. A User's Guide & Reference Manual. Springer-Verlag. 381pp.

BÜRER, H. (1991): Problems in estimation of phaeophytin. Verein. Internat. Verin. Limnol., 24:1259.

BURNS, D.M., MacDONALD, S.G.G. (1975) Physics for Biology and Pre-medical Students. 2nd Edition. Addison-Wesley Publishers Ltd. 665pp.

BURNS, J.C., COY, J.S., TERVET, J.D., HARRIMAN, R., MORRISON, B.R.S., QUINE, C.P. (1984): The Loch Dee Project: a study on the ecological effects of acid precipitation and forest management on an upland catchment in South West Scotland. 1. Preliminary investigations. Fisheries Management, 15(4):145-167.

BURROWS, E.M. (1987): Seaweeds of the British Isles Volume 2. Chlorophyta. British Museum, London. 238pp.

CHAMBERS, P.A., SPENCE, D.H.N. (1984): Diurnal changes in the ratio of underwater red to far red light in relation to aquatic plant photoperiodism. Journal of Ecology, 72:495-503.

CHRISMAN, T.L., SCHULZE, R.L., BREZONICK, P.L., BLOOM, S.A. (1980): Acid precipitation: the biotic response in Florida lakes. In: Ecological Impact of Acid Precipitation. Ed: D. Drabløs, A. Tollan. SNSF-project. Oslo-As. pp296-297.

CLAPHAM, A.R., TUTIN, T.G., MOORE, D.M. (1987): Flora of the British Isles. 3rd edition. Cambridge University Press, Cambridge. 688pp.

COLLINS, C.D., SHELDON, R.B., BOYLEN, C.B. (1987): Littoral zone macrophyte community structure and association of species along physical gradients in Lake George, New York, USA. Aquatic Botany, 29:177-194.

CONWAY, H.L., HENDREY, G.R. (1982): Ecological effects of acid precipitation on primary producers. Pp 179-195 in Acid Precipitation: Effects on Ecological Systems. Ed: F.M. D'itri. Ann Arbour Science, The Butterworth Group.

CRESSER, M.S., HARRIMAN, R., PUGH, K. (1988): Processes of acidification in soils and freshwater. Pp55-70 In: Acid Rain and Britain's Natural Ecosystems. Ed: M. Ashmore, N. Bell, C. Garrety. Imperial College Centre for Environmental Technology.

DELISLE, C.E., ROY, L., BILODEAU, P., ANDRÉ, P. (1984): Effects d'une acidification artificelle in situ sur le phytoplancton et le zooplankton lacustre. Verein. Internat. Verein. Limnol., 22:383-387.

DEN HARTOG, C. & SEGAL, C. (1964): A new classification of water plant communities. Acta Botanica Neerlandica, 13:367-393.

DICKINSON, G. (1991): Recreational impacts. Loch Lomond 1991. Proceedings from a symposium on environmental conditions in the Loch Lomond basin and their implications for resource management and development. Universities of Glasgow & Strathclyde. pp39-51.

DUARTE, C.M. (1991): Seagrass depth limits. Aquatic Botany, 40:363-377.

DUNCAN, W.G., LOOMIS, R.S., WILLIAMS, W.A., HANAN, R. (1967): A model for simulating photosynthesis in plant communities. Hilgardia, 38:181-205.

EKMAN, S. (1911): Neue apparate zur qualitativen und quantitativen erforschung der bodenfauna der seen. Int. Rev, Ges. Hydrobiol. Hydrogr., 3:553-561.

EHLERINGER, J.R., OSMOND, C.B. (1989): Stable Isotopes. In: Plant Physiology Ecology Field Methods and Instrumentation. Ed: R.W. Pearcy, J. Ehleringer, H.A. Mooney, P.W. Rundel. Chapman & Hall, New York. pp281-300.

EHLERINGER, J.R., RUNDEL, P.W. (1988): Stable isotopes: History, units, and instrumentation. In: Stable Isotopes in Ecological Research. Ecological Studies, Analysis & Synthesis no.68. Ed: P.W. Rundel, J.R. Ehleringer, K.A. Nagy. Spreinger-Verlag. New York. Pp 1-15.

EHLERINGER, J.R., SAGE, R.F., FLANAGAN, L.B., PEARCY, R.W. (1991): Climate change and the evolution of C4 photosynthesis. Trends in Ecology and Evolution, 6(3):95-99.

ERIKSSON, F., HÖRNSTRÖM, E., MOSSBERG, P., NYBERG, P. (1983): Ecological effects of lime treatment of acidified lakes and rivers in Sweden. Hydrobiologia, 101:145-164.

FARMER, A.M. (1987): Photosynthetic adaptation in *Lobelia dortmanna* and other isoetids. Photosynthetica, 21(1):51-55.

FARMER, A.M. (1990): The effects of lake acidification an aquatic macrophytes - a review. Environmental Pollution, 65:219-240.

FARMER, A.M, ADAMS, M.S. (1989): A consideration of the problems of scale in the study of the ecology of aquatic macrophytes. Aquatic Botany, 33:177-189.

FARMER, A.M., SPENCE, D.H.N. (1986): The growth strategies and distributions of isoetids in Scottish freshwater lochs. Aquatic Botany, 26:247-258.

FARMER, A.M. & SPENCE, D.H.N. (1987): Environmental control of the seasonal growth of the submerged macrophyte *Lobelia dortmanna* L. New Phytologist, 106:289-299.

FARQUHAR, G.D., EHLERINGER, J.R., HUBIK, K.T. (1989): Carbon isotope discrimination and photosynthesis. Annual Review Plant Physiology and Plant Molecular Biology, 40:503-537.

FOZZARD, I., MARSDEN, M. (1985): Lake of Menteith Surveys 1981-1985. Unpublished report to the Nature Conservancy Council.

GACIA, E., BALLESTEROS, E. (1993): Population and individual variability of *Isoetes lacustris* L. with depth in a Pyrenean lake. Aquatic Botany, 46:35-47.

GAUCH, H.G. (1986): Multivariate Analysis in Community Ecology. Cambridge University Press. 298pp.

GOLTERMAN, H.L. (1975): Developments in Water Science 2. Physiological Limnology. Elsevier Scientific Publishing, Amsterdam, 489pp.

GRAHN, O. (1985): Macrophyte biomass and production in Lake Gårdsjön - an acidified clearwater lake in SW Sweden. Ecological Bulletins, 37:203-212.

GRIME, J.P., CRICK, J.C., RINCON, J.E. (1986): The ecological significance of plasticity. In: Symposia for the Society of Experimental Botany no.40 Plasticity in Plants. Ed: A.J. Trewavas, D.H. Jenning. Society for Experimental Botany, Great Britain. Pp5-30.

GUPPIE, H.B. (1917): Plants, Seeds & Currents in the Azores. London. 531pp.

HANDLEY, L.L., DAFT, M.J., WILSON, J., SCRIMGEOUR, C.M., INGLEBY, K., SATTAR, M.A. (1993): Effects of the ecto- and VA-mycorrhizal fungi *Hydnagium carneum* and *Glomus clarum* on the $\delta^{15}N$ and $\delta^{13}C$ values of *Eucalytus globulus* and *Ricinus communis*. Plant, Cell & Environment, 16:175-382.

HAMILTON, J.D. (1988): Recent influences on the ecology of Loch Lomond, Scotland. Verh. Internat. Verein. Limnol., 23:403-413.

HARPER, D.M. (1978): Limnological studies on three Scottish Lowland freshwater lochs. Unpublished PhD Thesis, University of Dundee.

HARRIMAN, R., MORRISON, B. (1982): Ecology of streams draining forested and non-forested catchments in an area of central Scotland subject to acid precipitaion. Hydrobiologia, 88:251-263.

HASLER, A.D., JONES, E. (1949): Demonstration of the antagonistic action of large aquatic plants on algae and rotifers. Ecology, 30(30):359-365.

HENDRY, G.R., VERTUCCI, F.A. (1980): Benthic plant communities in acidic Lake Colder, New York: *Sphagnum* and the algal mat. Ecological effect of acid precipitation. Proceedings of an International Conference, Sandefjord, Norway. Ed: D. Drabløs, A. Tolla. Pp314-315.

HILL, M.O., GAUCH, H.G. (1980): Detrended Correspondence Analysis, an improved ordination technique. Vegetatio, 42:47-58.

HILMER, T., BATES, G.C. (1989): Filter types, filtration and post-filtration treatment in phytoplankton production studies. Journal of Plankton Research, 11(1):49-63.

HIPKINS, M.F., BAKER, N.R. (1986): Spectroscopy. In: Photosynthetic Energy Transduction. A Practical Approach. Ed: M.F. Hipkins & N.R. Baker. IRL Press, Oxford. 199pp.

HODGSON, J.M. (Ed) (1974): Soil Survey Field Handbook. Soil Survey Technical Monograph no.5. Harpenden. 90pp.

HOLM-HANSEN, O. & REIMAN, B. (1978): Chlorophyll *a* determination: improvements in methodology, Oikos, 30:438-447.

HOLSTRUP, O. & WIEGLEB, G. (1991a): The influence of different CO_2 concentrations in the light and the dark on diurnal malate rhythm and phosphoenolpyruvate carboxylase activities in leaves of *Littorella uniflora* (L.) Aschers. Aquatic Botany, 40:91-100.

HOLSTRUP, O. & WIEGLEB, G. (1991b): Anatomy of leaves of submerged and emergent forms of *Littorella uniflora* (L.) Ascherson. Aquatic Botany, 39:195-209.

HOOTSMANS, M.J.M. (1991): Allelopathic limitation of algal growth by macrophytes. In: Macrophytes, a key to understanding changes caused by eutrophication in shallow freshwater ecosystems M.J.M. Hootsmans & J.E. Vermaat. International Institute for Hydraulic and Environmental Engineering. The Netherlands. Pp131-156.

HOOTSMANS, M.J.M., VERMAAT, J.E. (1991): Light response curves of *Potamogeton pectinatus* L. as a function of plant age and irradiance level during growth. Chapt 4 In: Macrophytes a key to understanding changes caused by eutrophication in shallow freshwater ecosystems. M.J.M. Hootsmans & J.E. Vermaat. International Institute for Hydraulic and Environmental Engineering. The Netherlands. Pp57-130.

HOUGH, R.E., FORNWALL, H.D., NEGELE, B.J., THOMPSON, R.L. & PUTT, D., (1989): Plant community dynamics in a chain of lakes: principal factors in the decline of rooted macrophytes with eutrophication. Hydrobiologia, 173:199-217.

HOWARD-WILLIAMS, C. (1981): Studies on the ability of a *Potamogeton pectinatus* community to remove dissolved nitrogen and phosphorous from lake water. Journal of Applied Ecology, 18: 619-637.

HUNT, R. (1990): Plant Growth Analysis. Edward Arnold, London. 115pp.

HUTCHINGS, M.J. (1986) Plant population biology. Chapter 8 in: Methods in Plant Ecology. 2nd edition. Eds: P.D. Moore, S.B. Chapman. Blackwell Scientific Publications pp377-436.

ILMAVITRA, V. (1988): Acidic Lake Lakeenjärvi, Eastern Finland: phytoplankton succession and water chemistry. Verein. Internat. Verein. Limnol., 23:693-698.

IRVINE, K., MOSS, B., STANSFIELD, J. (1990): The potential of artificial refugia for maintaining a community of large-bodied cladocera against fish predation in a shallow eutrophic lake. Hydrobiologia 201:379-389.

JACKSON, H.O., STARRET, W.C., (1959): Turbidity and sedimentation at Lake Chautauqua, Illinois. Journal of Wildlife Management, 23:157-168.

JAMES, V. (1988) Notes on the search for *Najas flexilis* in Loch of Lowes Dunkeld, 1987, with an update for 1988, and preliminary plant list. Unpublished notes to Scottish Wildlife Trust, Loch of Lowes. 3pp.

JASSBY, A.D., PLATT, T. (1976): Mathematical formulation of the relationshipe between photosynthesis and ligh for phytoplankton. Limnology & Oceanography, 21:540-547.

JEFFRIES, M., MILLS, D. (1990): Freshwater Ecology. Principles and Applications. Belhaven Press, London & New York. 285pp.

JONES, C.R., WALTI, K., ADAMS, M.S. (1983): Phytoplankton as a factor in the decline of the submersed macrophyte *Myriophyllum spicatum* L. in Lake Wingra, Wisconsin. USA.

JONES, J.I., EATON, J.W., HARDWICK, K. (1993): Physiological plasticity in *Elodea nuttallii* (Planch.) St John. Journal of Aquatic Plant Management 31:88-94.

JUPP, B.P., SPENCE, D.H.N. (1977): Limitations on macrophytes in a eutrophic lake - Loch Leven 1 Effects of Phytoplankton. Journal of Ecology, 65:175-186.

KEDDY, P.A. (1982): Quantifying within lake gradients of wave energy: Inter-relationships of wave energy, substrate particle size and shorelins plants in Axe Lake, Ontario. Aquatic Botany, 14:41-58.

KEDDY, P.A. (1983): Shoreline vegetation in Axe Lake, Ontario: Effects of exposure on zonation patterns. Ecology, 64:331-334.

KEELEY, J.E. (1988): Stable carbon isotopes in vernal pool aquatics of differing photosynthetic pathways. In: Stable Isotopes in Ecological Research. Ecological Studies. Analysis & Synthesis no.68. Ed: P.W. Rundel, J.R. Ehleringer, K.A. Nagy. Spreinger-Verlag. New York. Pp 76-81.

KEELEY, J.E., STERNBERG, L.O., DENIRO, M.J. (1986): The use stable isotopes in the study of photosynthesis in freshwater plants. Aquatic Botany, 213-223.

KILMER, V.J., ALEXANDER, L.T. (1949): Methods for mechanical analysis of soils. Soil Science, 68:15.

KIRK, J.T.O (1975a): A theoretical analysis of the contribution of algal cells to the attenuation of light within natural waters I. General treatments of suspensions of pigmented cells. New Phytologist, 75:11-20.

KIRK, J.T.O (1975b): A theoretical analysis of the contribution of algal cells to the attenuation of light within natural waters II. Spherical cells. New Phytologist, 75:21-36.

KIRK, J.T.O (1976): A theoretical analysis of the contribution of algal cells to the attenuation of light within natural waters III. Cylindrical and spheroidal cells. New Phytologist, 77:341-358.

KIRK, J.T.O. (1983): Light and Photosynthesis in Aquatic Systems. Cambridge University Press. 401pp.

LACHAVANNE, J.-B. (1985): The influence of accelerated eutrophication on the macrophytes of Swiss lakes: Abundance and distribution. Verh. Int. Verein. Limnol., 22:2950-2955.

LAWRIE, C.L. (1902): Flowering Plants Their Structure and Habit. Allman & Sons, London.

LAZAREK, S. (1985): Epiphytic algal production in acidified Lake Gårdsjön, SW Sweden. Ecological Bulletin, 37:213-218.

LAZAREK, S. (1986): Responses of the *Lobelia*-epiphyte complex to liming of an acidified lake. Aquatic Botany, 25:73-81.

LEES, F.M. (1991): A Summary Report of Water Chemistry and Hydrology of Loch Dee, 1987-1989. Loch Dee Project Working Paper, June 1991. 46pp.

LEIVESTAD, H., HENDRY, G., MUNIZ, I.P., SNEKVIK, E. (1976): Effects of acid precipitation on freshwater organisms. Chapter 4 in: Impact of Acid Precipitation on Forest and Freshwater Ecosystems in Norway. Ed: F.H. Braekke. Summary report on research results of phase I (1972-1975) of the SNSF project NISK, 1432 Aas, Norway.

LENZ, J., FRITSCHE, P. (1980): The estimation of chlorophyll *a* in water samples. A comparative study on retention in a glass-fibre and membrane filters and on the reliability of two storage methods. Ergeb. Limnol., 14:46-51.

LOCH DEE PROJECT (1989): The Loch Dee Project: A study of catchment acidification: Summary report. Civil Engineering and water services Scottish Development Department.

LOCH DEE PROJECT (1993): Proceeding of the Loch Dee Symposium. Acidification, Forestry & Fisheries Management in Upland Galloway. Foundation for Water Research. 172pp.

LIPKIN, Y., BEER, S., BEST, E.P.H., KAIRESALO, T., SALONEN, K. (1986): Primary production of macrophytes: terminology, approaches and a comparison of methods. Aquatic Botany, 26:129-142.

LYDEN, A., GRAHN, O. (1985): Phytoplankton species composition and production in Lake Gårdsjön - an acidified clearwater lake in SW Sweden. Ecological Bulletins, 37:195-202.

MABERLY, S.C., SPENCE, D.H.N. (1983): Photosynthetic inorganic carbon use by by freshwater plants. Journal of Ecology, 71:705-724.

MADSEN, T.V. (1987a): Sources of inorganic carbon aquired through CAM in *Littorella uniflora* (L.) Aschers. Journal of Experimental Botany, 38:367-377.

MADSEN, T.V. (1987b): The effect of growth conditions on dark and light carbon assimilation in *Littorella uniflora.* Physiol. Plant, 70:183-188.

MADSEN T.V., SAND-JENSEN, K., BEER, S . (1993): Comparison of photosynthetic performance and carboxylation capacity in a range of aquatic macrophytes of different growth forms. Aquatic Botany, 44:373-384.

MAITLAND, P.S. (1981): Monographiae Biologicae Volume 44. The Ecology of Scotland's Largest Lochs. Lomond, Awe, Ness, Morar and Shiel. Dr W. Junk Publishers. 290pp.

MARKER, A.F.H., JINKS, S. (1982): The spectrophotometric analysis of chlorophyll a phaeopigments in acetone, ethanol and methanol. Hydrobiol. Beih. Ergebn. Limnol., 16:3-17.

MARRS, S.J., MURPHY, K.J, HILLS, J.M. (1993): The aquatic flora of Loch Dee, 1904-1990. In: Proceeding of the Loch Dee Symposium. Acidification, Forestry and Fisheries in Upland Galloway. Foundation for Water Research. Pp 97-106.

MARSDEN, R. (1990): Loch Dee Geology. Unpublished report to the 11th annual meeting of the Loch Dee Research Group.

MAULOOD, B.K., BONEY, A.D. (1979): A seasonal ecological study of the phytoplankton of Loch Lomond. Hydrobiologia, 71:239-259.

MAULOOD, B.K., BONEY, A.D. (1981): Phytoplankton ecology of the Lake of Menteith, Scotland. Hydrobiologia, 79:179-186.

MISRA, R.D. (1938): Edaphic factors in the distribution of aquatic plants in the English lakes. Journal of Ecology, 26:411-451.

MITCHELL, G. (1991): Water Management. In: Loch Lomond 1991. Proceedings from a symposium on environmental conditions in the Loch Lomond basin and their implications for resource management and development. Universities of Glasgow and Strathclyde. Pp 52-62.

MOELLER, R.E. (1975): Hydrophyte biomass and community structure in a small, oligotrophic New Hampshire lake. Verh. int. Ver. Limnol., 19:1004-1012.

MOELLER, R.E. (1978): Seasonal changes in biomass, tissue chemistry and net production of the evergreen *Lobelia dortmanna* L. Canadian Journal of Botany, 56:1425-1433.

MOORE, J.A. (1986): Charophytes of Great Britain & Ireland B.S.B.I. Handbook No.5. Botanical Society of the British Isles. 140pp.

MOORE, P.D., CHAPMAN, S.B. Eds. (1986): Methods in Plant Ecology. 2nd Edition. Blackwell Scientific Publications. 589pp

MORRIS, J.T. (1991): Effects of nitrogen loading on wetland ecosystems with particular reference to atmospheric deposition. Annual Review of Ecology and Systematics, 22:257-279.

MORRISON, B.R.S., BATTARBEE, R.W. (1988): Acid Rain. Effects on freshwater flora and fauna (excluding fish). In: Acid Rain and Britain's Natural Ecosystems. Imperial College Centre for Environmental Technology. Pp 71-86.

MOSS, B., STANSFIELD, J., IRVINE, K. (1991): Development of daphnid communities in diatom- and cyanophyte-dominated lakes and their relevance to lake restoration by biomanipulation. Journal of Applied Ecology, 28:586-602.

MULLIGAN, H.F., BARANOWSKI, A. (1969): Growth of phytoplankton and vascular plants at different nutrient levels. Verh. Int. Verein. Limnol., 17:802-810.

MULLIGAN, H., BARANOWSKI, A., JOHNSON, R. (1976): Nitrogen and phosphorous fertilisation of aquatic vascular plants and algae in replicated ponds 1. Initial response to fertilisation. Hydrobiologia, 44(2):109-116.

MURPHY, K.J., MILLER, S., ANDERSON, K. (1986): Aquatic vegetation of Loch Dee, Galloway. Unpublished report to DAFS Freshwater Fisheries Laboratory, Pitlochry.

NALEWJKO, C., PAUL, B. (1985): Effects of manipulations of aluminium concentration and pH on phosphate uptake and photosynthesis of planktonic communities in two Precambrian shield lakes. Canadian Journal of Fisheries and Aquatic Science, 42:1946-1953.

NATURE CONSERVANCY COUNCIL (1985): Lochs of Butterstone, Craiglush and Lowes SSSI. Unpublished records of NCC Perth & Kinross District, Tayside Region.

NELSON, D.W., SUMMERS, L.E. (1973): Determination of total nitrogen in plant material. Agronomy Journal, 65:109-112.

NEUVONEN, S., SUOMELA, J. (1990): The effects of simulated acid rain on pine needle and birch litter decomposition. Journal of Applied Ecology, 27:857-872.

NEWBOLD, C., PALMER, M. (undated): *Elatine hydropiper* - a recent record for the Lake of Menteith (VC 87). Unpublished records of Nature Conservancy Council for Scotland, South-East.

NIELSEN, S.L., SAND-JENSEN, K. (1991): Variation in growth rates of submerged rooted macrophytes. Aquatic Botany, 39:109-120.

NOBEL, P.S. (1991): Tansley Review No. 32. Achievable productivities of certain CAM plants: basis for high values compared with C3 and C4 plants. New Phytologist, 119:183-205.

NOBEL, P.S., FORSETH, I.N., LONG, S.P. (1993): Canopy structure & light interception. Chapter 6 in: Photosynthesis and Production in a Changing Environment: A Field & Laboratory Manual. Ed: D.O. Hall, J.M.O. Scurlock, H.R. Bolhár-Nordenkampf, R.C. Leegood, S.P. Long. Chapman & Hall, London. Pp 79-90.

ORDNANCE SURVEY (1973): Sheet NN59NE 1:10,000 series.

ORDNANCE SURVEY (1977a): Sheet NX47NE 1:10,000 series.

ORDNANCE SURVEY (1977b): Sheet NS39SE 1:10,000 series.

ORDNANCE SURVEY (1978a): Sheet NN50SE 1:10,000 series.

ORDNANCE SURVEY (1978b): Sheet NO04SE 1:10,000 series.

ORDNANCE SURVEY (1983): Sheet NO04SW 1:10,000 series.

ORDNANCE SURVEY (1981): Sheet NX47/57, Pathfinder series 527.

ORDNANCE SURVEY (1983): Sheet NO04SW 1:10,000 series.

OSMOND, C.B., VALAANE, N., HASLAM, S.M., UOTILA, P., ROKSANDIC, Z. (1981): Comparisons of $\delta^{13}C$ values in leaves of aquatic macrophytes from different habitats in Britain and Finland; some implications for photosynthesis processes in aquatic plants. Oecologia, 50:117-124.

OZIMEK, T., PIECZYÑSKA, E., HANKIEWICZ, A. (1991): Effects of filamentous algae on submerged macrophyte growth: a laboratory experiment. Aquatic Botany, 41:309-315.

PEARCY, R.W. (1990): Sunflecks and photosynthesis in plant canopies. Annual Review of Plant Physiology & Plant Molecular Biology, 41:421-453.

PEARSALL, W.H. (1918): On the classification of aquatic plant communities. Journal of Ecology, 6:75-84.

PEDERSEN, O. (1993): Long distance water transport in aquatic plants. Plant Physiology, 103:1369-1375.

PEDERSEN, O., SAND-JENSEN, K. (1993): Water transport in submerged macrophytes. Aquatic Botany, 44:385-406.

PEÑUELAS, J. (1988): An experimental system to study the effects of pressure, light and temperature on macrophyte production. Pol. Arch. Hydrobiol., 35(1):17-21.

PIZARRO, G.M. MONTECINO, V. (1992): Primary productivity and maintenance through chlorophyll *a* changes in *Elodea potamogeton* through an annual cycle. Aquatic Botany, 43:231-248.

PHILIPS, G.L., EMINSON, D., MOSS, B. (1978): A mechanism to account for macrophyte decline in progressively eutrophicated freshwaters. Aquatic Botany, 4:103-126.

PRESCOTT, G.W. (1970): How to Know the Freshwater Algae. WmC Brown Company Publishers. Dubuque, Iowa. 348pp.

PRESTON, C.D. (1987): Kindrogen Field Centre, Perthshire 20-27th September. Botanical Society for the British Isles News, 46:45-46.

RAVEN, J.A. (1970): Exogenous inorganic carbon sources in plant photosynthesis. Biological Reviews, 45:167-221.

RAVEN, J.A., HANDLEY, L.L., MACFARLANE, J.J., McINROY, S., McKENZIE, L., RICHARDS, J.H., SAMUELSON, G. (1988): Tansley Review No. 13. The role of CO_2 uptake by roots and CAM in the acquisition of inorganic C by plants of the isoetid life-form: a review, with new data on *Eriocaulon decangulare* L. New Phytologist, 108:125-148.

RAVEN, J.A., MacFARLANE, J.J., GRIFFITHS, H. (1987): The application of carbon isotope discrimination techniques. In: Plant Life in Aquatic and Amphibious Habitats. Ed: R.M.M. Crawford. Special Publication no.5 of the British Ecological Society. pp129-149.

RAVEN, J.A., OSBORNE, B.A., JOHNSTON, A.M. (1985): Uptake of carbon dioxide by aquatic vegetation. Cell and Environment, 8:417-425.

RAVEN, P.J. (1988): Occurrence of *Sphagnum* moss in the sublittoral of several small oligotrophic lakes in Galloway, south-west Scotland. Aquatic Botany, 30:223-230.

REIMAN, B. (1980): A note on the use of methanol as an extraction solvent chlorophyll determination. Ergeb. Limnol., 14:70-78.

REIMAN, B., ERNST, D. (1982): Extraction of chlorophylls *a* and *b* from phytoplankton using standard extraction techniques. Freshwater Biology, 12:217-223.

ROBE, W.E., GRIFFITHS, H. (1988): C3 and CAM photosynthetic characteristics of the submerged macrophyte *Littorella uniflora*: Regulation of leaf internal CO_2 supply in response to variation in rooting substrate inorganic carbon concentration. Journal of Experimental Botany, 39:1397-1410.

ROBE, W.E., GRIFFITHS, H. (1990): Photosynthesis of *Littorella uniflora* grown under two PAR regimes: C3 and CAM gas exchange and the regulation of internal CO_2 and O_2 concentrations. Oecologia, 65:128-136.

ROBE, W.E., GRIFFITHS, H. (1992): Seasonal variation in the ecophysiology of *Littorella uniflora* L. Ascherson. New Phytologist, 120(2): 289-304.

ROBSON, T.O. (1974): New techniques for aquatic weed research. Chemistry & Industry 20 July 1974: 584-9.

ROELOFS, J.G.M. (1983): Impact of acidification and eutrophication on macrophyte communities in soft waters in The Netherlands 1. Field observations. Aquatic Botany, 17:139-155.

ROELOFS, J.G.M., SCHUURKES, J.A.A.R. (1983): Impact of acidification and eutrophication on macrophyte communities in soft waters. Proceeding of International Conference on Aquatic Macrophytes, Nijmegen, 197-202.

ROELOFS, J.G.M., SCHUURKES, J.A.A.R., SMITS, A.J.M. (1984): Impact of acidification and eutrophication on macrophyte communities in soft waters. II. Experimental studies. Aquatic Botany, 18:389-411

RØRSLETT, B. (1991): Principal determinants of aquatic macrophyte richness in north European lakes. Aquatic Botany, 39:173-193.

SALVUCCI, M.E., BOWES, G. (1983): Two photosynthetic mechanisms mediating the low photorespiratory state in submersed aquatic angiosperms. Plant Physiology, 73:488-496.

SAND-JENSEN, K. (1977): Effects of epiphytes on Eelgrass photosynthesis. Aquatic Botany, 3:55-63.

SAND-JENSEN, K. (1978): Metabolic adaptation and vertical zonation of *Littorella uniflora* (L.) Aschers. and *Isoetes lacustris* L. Aquatic Botany, 4:1-10.

SAND-JENSEN, K. (1983): Photosynthetic carbon sources of stream macrophytes. Journal of Experimental Botany, 34:198-210.

SAND-JENSEN, K. (1987): Environmental control of bicarbonate use among freshwater and marine macrophytes. In: Plant Life in Aquatic and Amphibious Habitats. Ed: R.M.M. Crawford. Special Publication number 5, British Ecological Society. Blackwell Scientific Publications. 452pp.

SAND-JENSEN, K. & SØNDERGAARD, M. (1978): Growth and production of isoetids in oligotrophic Lake Kalgaard, Denmark. Verh. Int. Verein. Limnol., 20:659-666.

SAND-JENSEN K. & SØNDERGAARD, M. (1979): Distribution and quantitative development of aquatic macrophytes in relation to sediment characteristics in oligotrophic Lake Kalgaard, Denmark. Freshwater Biology, 9:1-11.

SAND-JENSEN K. & SØNDERGAARD, M. (1981): Phytoplankton and epiphyte development and their shading effect on submerged macrophytes in lakes of different nutrient status. Int. Revue Ges. Hydrobiol., 66(4):529-552.

SCHANZ, F., FELIX, V. (1991): Phytoplankton development in Lake Zurich and changes in the vertical attenuation coefficient (K_d), the contributions to gilvin (K_G), phytoplankton (K_{Ph}) and tripton (K_{TR}) during spring 1986. Verh. Internat. Verein. Limnol., 24:949-954.

SCHEFFER, M. (1990): Multiplicity of stable states in freshwater systems. Hydrobiologia, 200/201:475-486.

SCHEFFER, M. (1991): Fish and nutrients interplay determines algal biomass: a minimal model. Oikos 62:271-282.

SCHOENBERG, S.A. (1990): Short-term productivity responses of algae and bacteria to zooplankton grazing in two freshwater lakes. Freshwater Biology, 23:395-410.

SCULTHORPE, C.D. (1967): The Ecology of Aquatic Vascular Plants. Arnold, London. 610pp.

SEDDON, B. (1972): Aquatic macrophytes as limnological indicators. Freshwater Biology, 2:107-130.

SHELDON, R.B., BOYLEN, C.W. (1978): An underwater survey method for estimating submerged macrophyte population biomass and density. Aquatic Botany, 4:65-72

SIMPSON, D.A. (1984): A short history of the spread of *Elodea* Michx in the British Isles. Watsonia, 15:1-19.

SIMPSON, P.S., EATON, J.W., HARDWICK, K. (1980): The influence of environmental factors on apparent photosynthsis and respiration of the submerged macrophyte *Elodea canadensis*. Plant, Cell & Environment, 3:415-423.

SIMPSON, P.S., EATON, J.W. (1986): Comparative studies of the photosynthesis of the submerged macrophyte *Elodea canadensis* and the filamentous algae *Cladophora glomerata* and *Spirogyra* sp. Aquatic Botany, 24:1-12.

SLACK, H.D. (1957): Studies on Loch Lomond 1. Blackie & Son, Ltd. 132pp.

SMITH, A.J.E. (1978): The Moss Flora of Britain and Ireland Cambridge University Press. 706pp.

SMITH, F.A., WALKER, N.A. (1980): Photosynthesis by aquatic plants: Effects of unstirred layers in relation to assimilation of CO_2 and HCO_3^- and to carbon isotope discrimination. New Phytologist, 86:245-259.

SMITH, R.A.H., STEWART, N.F., TAYLOR, N.W., THOMAS, R.E. (1992): A Checklist of the Plants of Perthshire. Perthshire Society of Natural Science. 69pp.

SMITS, A.J.M., LAAN, P., THIER, R.H., VAN DER VELDE, G. (1990): Root aerenchyma, oxygen leakage patterns and alcoholic fermentation ability of the roots of some nymphaeid and isoetid macrophytes in relation to the sediment type of their habitat. Aquatic Botany, 38:3-17.

SOKAL, R.R., ROHLF, F.J. (1981): Biometry. W.H. Freeman and Co., New York. 859pp.

SØNDERGAARD, M. (1979): Light and dark respiration and the effect of the lacunal system on refixation of CO_2 in submerged aquatic plants. Aquatic Botany, 6:269-283.

SØNDERGAARD, M. (1988): Photosynthesis of aquatic plants under natural conditions. In: Hanbook of Vegetation Science: Vegetation of Inland Waters. Ed: J.J. Symoens. Kluwer Academic Publishers. 385pp.

SØNDERGAARD, M. & BONDE, G. (1988): Photosynthetic characteristics and pigment content and composition in *Littorella uniflora*(L.) Aschers. in a depth gradient. Aquatic Botany, 32:307-319.

SØNDERGAARD, M. & SAND-JENSEN, K. (1979): Carbon uptake by leaves and roots of *Littorella uniflora* L. (Aschers.). Aquatic Botany, 6:1-12.

SPENCE, D.H.N. (1964): The macrophytic vegetation of freshwater lochs, swamps and associated fens. In: The Vegetation of Scotland Ed: J.H. Burnett. Oliver & Boyd, Edinburgh, pp306-425.

SPENCE, D.H.N. (1967): Factors controlling the distribution of freshwater macrophytes with particular reference to the lochs of Scotland. Journal of Ecology, 55:147-170.

SPENCE, D.H.N. (1982): The zonation of plants in freshwater lakes. Advances in Ecological Research, 12:37-125.

SPENCE, D.H.N., BARCLAY, A.M., ALLEN, E.D. (1979): Limnology and macrophyte vegetation of a deep clear limestone lake, Lake Borralie. Transactions of the Royal Society of Edinburgh, 44:187-204.

SPENCE, D.H.N., CAMPBELL, R.M., CHRYSTAL, J. (1973): Specific leaf areas and zonation of freshwater macrophytes. Journal of Ecology, 61:317-328.

SPENCE, D.H.N., CHRYSTAL, J. (1970a) Photosynthesis and zonation of freshwater macrophytes 1: Depth distribution and shade tolerance. New Phytologist, 69:205-215.

SPENCE, D.H.N., CHRYSTAL. J. (1970b): Photosynthesis and zonation of freshwater amcrophytes. II: Adaptability of species of deep and shallow water. New Phytologist, 69:219.

SPENCER & BOWES, G. (1990): Ecophysiology of the world's most troublesome aquatic weeds. Chapter 4 in Aquatic Weeds: The Ecology and Management of Nuisance Aquatic Vegetation. Eds: A.H. Pieterse, K.J. Murphy. Oxford Science Publications. 593pp.

SPINK, A.J. (1992): The Ecological Strategies of Aquatic *Ranunculus* species. PhD Thesis, University of Glasgow.

STACE, C. (1991): New Flora of the British Isles Cambridge University Press. 1226 pp.

STANSFIELD, J., MOSS, B., IRVINE, K. (1989): The loss of submerged macrophytes with eutrophication III. Potential role of organochlorine persticides: a palaeoecological study. Freshwater Biology, 22:109-132.

STEVENSON, R.L., LYON, G.L. (1982): Carbon-13 depletion in an estuarine bivalve: detection of marine and terrestrial food sources. Oecologia, 55:100-113.

STRYER, L. (1981): Biochemistry 2nd Edition. W.H. Freeman and Company, New York. 949pp.

SWARBRICK, J.T. & RAYMOND, J.C. (1970): The identification of the seeds of the British *Plantginaceae.* Annals of Botany, 34:1123-1129.

SZMEJA, J. (1987a): The ecology of *Lobelia dortmanna* L. I. The plasticity of individuals within a constant depth interval in oligotrophic lakes. Ekologia Polska, 35:497-522.

SZMEJA, J. (1987b): The ecology of *Lobelia dortmanna* L. III. The plasticity of individuals along a gradient of increasing depth in oligotrophic lakes. Ekologia Polska, 25:545-558.

SZMEJA, J (1987c): The structure of a population of *Lobelia dortmanna* L. along a gradient of increasing depth in an oligotrophic lake. Aquatic Botany, 28:1-13.

SZMEJA, J. (1989): Materials for the knowledge of the rare and endangered species of Poland 1. *Littorella uniflora* (L.) Aschers. (in Polish). Fragmenta Floristica et Geobotanica, 33:421-431.

TANNER, C.B., JACKSON, M.L. (1947): Nomographs of sedimentation times for soil particles under gravity or centrifugal acceleration. Proc. Soil Sci. Soc. Am., 12:60-65.

TENHUNEN, J.D., TENHUNEN, L.C., ZIEGLER, H., STICHLER, W., LANGE, O.L. (1982): Variation in carbon isotope ratios of Sempervivoide species from different habitats. Oecologia, 55:217-224.

TER BRAAK, C.J.F. (1986): Canonical Correspondance Analysis: A new eigenvector technique for multivariate direct gradient analysis. Ecology, 67:1167-1179.

TER BRAAK, C.J.F. (1987): Ordination. Chapter 5 in: Data Analysis in Community Landscape Ecology. Eds: R.H.G. Jongman, C.J.F. ter Braak, O.F.R. van Tongeren. Pudoc Wagenengen. pp91-173.

TER BRAAK, C.J.F. (1988): CANOCO - a FORTRAN programme for canonical community ordination by [partial] [detrended] [canonica] correspondence analysis, principal components analysis and redundancy analysis (version 2.1). Technical Report: LWA-88-02. Groep Landbounwwiskunde Wageningen, The Netherlands.

TERVET, D.J., HARRIMAN, R. (1988): Changes in pH and calcium after selective liming in the catchment of Loch Dee, a sensitive and rapid-tun-over loch in south-west Scotland. Aquaculture and Fisheries Management, 19:191-203.

TING, I.P. & RAYDOR, L. (1982): Regulation of C3 to CAM shifts. In: Crassulacean Acid Metabolism. Ed: I.P. Ting, M. Gibbs. American Society of Plants Physiologists, Rockville. 316pp.

TITUS, J.E., ADAMS, M.S. (1979): Coexistance and comparative light regimes of the submerged macrophytes *Myriophyllum spicatum* L. and *Vallisneria americana* Michx. Oceologia, 40:273-286.

TITUS, J.E., GOLDSTEIN, R.A., ADAMS, M.S., MANKIN, J.B., O'NEILL, R.V., WEILER, P.R., SHUGART, H.H., BOOTH, R.S.

(1975): A production model for *Myriophyllum spicatum* L. Ecology, 56:1129-1138.

TITUS, J.E., HOOVER, D.T. (1993): Reproduction in two submersed macrophytes declines progressively at low pH. Freshwater Biology, 30(1):63-72.

TOWNSEND, S.A., BOLAND, K.T., WRIGLEY, T.J. (1992): Factors contributing to a fish kill in the Australian wet dry tropics. Water Research, 26(8):1039-1044.

TRIFONOVA, I.S. (1989): Changes in community structure and productivity of phytoplankton as indicators of lake and reservoir eutrophication. Arch. Hydrobiol. Beih. Ergebn. Limnol., 33:363-371.

VAN DIJK, G.M., ACHTERBERG, E.P. (1993): Light climate in the water column of a shallow eutrophic lake (Lake Veluwe) in The Netherlands. Arch. Hydrobiol., (in press)

VAN VIERSSEN, W., PRINS, T.C. (1985): On the relationships between the growth of algae and aquatic macrophytes in brackish water. Aquatic Botany, 21:165-179.

VOËGE, M. (1989): Diving studies of isoetids in several acidic lakes of southern Norway. (In German). Tuexenia, 0(9);29-38.

WALKER, D.A. (1990): Use of oxygen electrode and fluorescence probes in simple measurements of photosynthesis. 2nd edition. Oxygraphics Ltd., Brighton/Packard Publishing, Chichester. 203pp.

WALKER, D.A. (1993): Polarographic measurement of oxygen. In: Photosynthesis and Production in a Changing Environment. A Field & Laboratory Manual. Ed: D.O.Hall, J.M.O., Scurlock, R.C. Leegood, S.P. Long. Chapman & Hall. pP168-180.

WEISNER, S.E.B. (1987): The relation between wave exposure and distribution of emergent vegetation in a eutrophic lake. Freshwater Biology, 18:537-544.

WELSH, W.T. & BURNS, J.C. (1987): The Loch Dee Project: runoff and surface water quality in an area subject to acid precipitation and afforestation in SW Scotland. Transactions of the Royal Society of Edinburgh, 78:249-260.

WERRITY, A., MAUCOTEL, S. (1993): Hydrochemistry of lake sediments and impact of liming in Loch Dee. In: Proceedings of the Loch Dee Symposium. Acidification, Forestry & Fisheries Management in Upland Galloway. Foundation for Water Research. Pp49-60.

WESTLAKE, D.F. (1978): Rapid exchange of oxygen between plant and water. Verh. Internat. Verein. Limnol., 20:2363-2367.

WETZEL, R.G. (1988): Water as an environment for plant life. In: Handbook of Vegetation Science Volume 15/1. Vegetation of Inland Waters. Ed: J.J. Symoens. Kluwer Academic Publishers, The Netherlands. Pp1-30.

WETZEL, R.G., HOUGH, R.A. (1973): Productivity and the role of aquatic macrophytes in lakes. Pol. Arch. Hydrobiol., 20:9-19.

WILSON, A. (1949): The altitudinal range of British plants. North Western Naturalist (supplement).

WILSON, S.D. (1991): Plasicity, morphology & distribution in twelve lakeshore plants. Oikos, 62:292-298.

WILSON, S.D. & KEDDY, P.A. (1985): Plant zonation on a shoreline gradient: physiological response curves of component species. Journal of Ecology, 72:185-200.

WIUM-ANDERSEN, S. (1987): Allelopathy among aquatic plants. Arch. Hydrobiol. Beih., 27:167-172.

WIUM-ANDERSEN, S., ANDERSEN, J.M. (1972): CO_2 content of the interstitial water in the sediment of Grane Langso, a Danish *Lobelia* lake. Limnology and Oceanography, 17:943-947.

WIUM-ANDERSEN, S., ANTHONI, U., CHRISTOPHEREN, C., HOVEN G. (1982): Allelopathic effects on phytoplankton by substances isolated from aquatic macrophytes (Charales). Oikos, 39:187-190.

WIUM-ANDERSEN, S. & BORUM, J. (1984): Biomass variation and autotrophic production od an epiphyte-macrophyte community in a coastal Danish area: eelgrass *Zostera marina* L. biomass and net production. Ophelia, 23:33-46.

WOODWARD, F.I. (1990): From ecosystems to genes: the importance of shade tolerance. Trends in Ecology & Evolution, 5:111-115.

YAN, N.D., STOKES, P.M. (1987): Phytoplankton of an acid lake and its' response to experimental alteration of pH. Environ. Cons. 5:93-100.

Appendices:

Appendix A: **Loch Surveys.**

During the autumn and winter of 1989/90 ten lochs were surveyed to determine their suitability for this study. Surveys were carried out by grapnel sampling either from the shore or from a small boat. In all cases species were recorded on a presence/absence basis. Some environmental data were recorded using the same field meters as in the 1990 field work - see Chapter 3.

The sites surveyed were as follow: Loch of Lowes in Perthshire; Loch na h'Achlaise and a small un-named loch to the east of Loch na h'Achlaise in Argyll; Loch Dee in Galloway; Linlithgow Loch in Lothian; two lochs in Strathclyde Park, Strathclyde, the main loch (called here Strathclyde) and Island Loch; Loch Rusky and Lake of Menteith near Aberfoyle; and two sites in Loch Lomond - the north site at Sloy Power Station and the second site in the Narrows between the islands of Inchtavannach and Inchmoan.

Data from these surveys are summarised in Tables A.1-A.3

Table A.1 Summary of Loch Survey sites.

Site	N.G.R.	Date	Survey Method
Lowes	NO 046 440	21 September	Small boat
h'Achlaise		4 October	Shore
No-name		4 October	Shore
Loch Dee	NX 470 970	11 October	Shore - snorkel
Linlithgow		19 October	Shore
Strathclyde		3 November	Small boat
Loch Rusky		14 November	Small boat
Lomond Narrows	NS 372 916	24 November	Small boat
Lomond North		5 January	Shore
Menteith	NN 572 007	11 January	Shore

Table A.2 Summary of environmental data collected

Site	pH	Cond (μScm^{-1})	DO (%/mgl^{-1})	E	Temp (°C)
Loch of Lowes	n/a	87	n/a	1.15	16
Loch na h'Achlaise	5.2	35	64/7.8	1.53	10
No-name	6.0	n/a	n/a	n/a	n/a
Loch Dee	n/a	37	75/9.2	1.11	11
Linlithgow	7.8	520	72/9.2	2.8	12
Strathclyde (Main)	n/a	n/a	n/a	n/a	n/a
Island Loch	7.5	446	58/7.5	n/a	9
Loch Rusky	7.0	77	54/7.3	2.7	7
Lomond (Narrows)	7.9	48	53/6.4	n/a	9
Lomond (North)	n/a	n/a	n/a	n/a	n/a
Menteith	n/a	n/a	n/a	n/a	n/a

Abbreviations used in Table A.2

Cond - conductivity
DO - dissolved oxygen
E - extinction coefficient
Temp - temperature
n/a - not available

Table A.3 Summary of plant species recorded

Site	*Species
Lowes	*Callitriche hermaphroditica, Elodea canadensis, Littorella uniflora, Lobelia dortmanna, Myriophyllum alterniflorum, Nitella opaca, Potamogeton gramineus, P. perfoliatus, P. pusillus*
h'Achlaise	*Carex* c.f. *lasiocarpa, Eleocharis acicularis, Equisetum fluviatile, Juncus bulbosus, Littorella uniflora, Lobelia dortmanna, Myriophyllum alterniflorum, Ranunculus flammula, Subularia aquatica, Utricularia vulgaris, Potamogeton natans*
No-name	*Carex* c.f. *lasiocarpa, Equisetum fluviatile, Juncus bulbosus, Menyanthes trifoliata, Myriophyllum alterniflorum, Potamogeton natans. P. polygonifolius, Utricularia vulgaris*
Dee	*Caltha palustris, Carex rostrata, Eleocharis palustris, Equisetum fluviatile, Juncus bulbosus, Isoetes lacustris, Littorella uniflora, Lobelia dortmanna, Myriophyllum alterniflorum, Ranunculus flammula, Subularia aquatica*

*for full species names and authorities see Appendix B.

Table A.3 continued

Site	Species
Linlithgow	*Callitriche hermaphroditica, Elodea canadensis, Glyceria maxima, Phalaris arundinacea, Potamogeton crispus, P. pectinatus*
Strathclyde (Main)	no macrophyte species found
Island Loch	*Elodea canadensis, Lemna trisulca, Potamogeton natans*
Rusky	*Callitriche* c.f. *hamulata, Carex* sp., *Elodea canadensis, Equisetum fluviatile, Isoetes lacustris, Littorella uniflora, Mentha aquatica, Myriophyllum spicatum, Potamogeton compressus, P. natans, Ranunculus flammula*
Lomond (Narrows)	*Eleocharis acicularis, Elodea canadensis, Eurhynchium praelongum, Hydrocotyle vulgaris, Isoetes lacustris, Juncus bulbosus, Littorella uniflora, Lobelia dortmanna, Myriophyllum alterniflorum, Potamogeton crispus, P. polygonifolius, Ranunculus flammula*
Lomond (Sloy)	*Agrostis stolonifera, Callitriche* c.f. *hamulata, Eleocharis acicularis, Fontinalis antipyretica, Juncus bulbosus, Littorella uniflora, Myriophyllum alterniflorum, Ranunculus flammula, R. sceleratus*

Menteith *Agrostis stolonifera, Isoetes lacustris, Juncus bulbosus, Littorella uniflora, Myosotis scorpioides, Phragmites australis, Ranunculus flammula, R. sceleratus,*

Appendix B:

Full names, authorities and common names for British plant species.

Nomenclature follows that of Stace, 1991 with the exception of charophytes which follow Moore, 1986 and mosses which follow Smith, 1978.

Agrostis stolonifera L., Creeping Bent
Callitriche hamulata Kuetz. ex Koch, Intermediate Water-starwort
Callitriche hermaphroditica L., Autumnal Water-starwort
Caltha palustris L. (*C. radicans* T.F. Forster) Marsh-marigold
Carex rostrata Stokes, Bottle Sedge
Chara globularis Thuill. var. *globularis*
Elatine hexandra (Lapeierre) DC., Six-stamened Waterwort
Elatine hydropiper L., Eight-stamened Waterwort
Eleocharis acicularis (L.) Roemer & Schultes, Needle Spike-rush
Elodea canadensis Michaux, Canadian Waterweed
Elodea nuttallii (Planchon) H. St John, Nuttall's Water-weed
Equisetum fluviatile L., Water Horsetail
Eurynchium praelongum (Hedw.) Br. Gur.
Fontinalis antipyretica Hedw.
Glyceria maxima (Hartman) O. Holmb., Reed Sweet-grass
Hydrocotyle vulgaris L., Marsh Pennywort
Isoetes lacustris L., Quillwort
Isoetes echinospora Durieu, Spring Quillwort
Juncus bulbosus L. (*J. kochii* F. Schultz), Bulbous Rush
Lemna trisulca L., Ivy-leaved Duckweed
Littorella uniflora (L.) Asch., Shoreweed
Lobelia dortmanna L., Water Lobelia
Mentha aquatica L., Water Mint
Menyanthes trifoliata L., Bogbean
Myosotis scorpioides L., Water Forget-me-not
Myrica gale L., Bog-myrtle
Myriophyllum alterniflorum DC., Alternate Water-milfoil
Myriophyllum spicatum L., Spiked Water-milfoil

Naja flexilis (Willd.) Rostkov & W. Schmidt, Slender Naiad
Nitella flexilis (L.) Agardh
Nitella opaca (Bruz.) Agardh
Nitella translucens (Persoon) Agardh
Phalaris arundinacea L., Reed Canary-grass
Phragmites australis (Cav.) Trin. ex Steudel
(*P. communis* Trin.), Common Reed
Potamogeton alpinus Balbis, Red Pondweed
Potamogeton compressus L., Grass-wrack Pondweed
Potamogeton crispus L., Curled Pondweed
Potamogeton filiformis Pers., Slender-leaved Pondweed
Potamogeton gramineus L., Various-leaved Pondweed
Potamogeton obtusifolius Mert. & Koch, Blunt-leaved Pondweed
Potamogeton natans L., Broad-leaved Pondweed
Potamogeton pectinatus L., Fennel Pondweed
Potamogeton perfoliatus L., Perfoliate Pondweed
Potamogeton polygonifolius Pourret, Bog Pondweed
Potamogeton pusillus L. (*P. panormitanus* Biv.), Lesser Pondweed
Ranunculus flammula L., Lesser Spearwort
Ranunculus petltatus Schrank, Pond Water-crowfoot
Ranunculus sceleratus L., Celery-leaved Buttercup
Schoenoplectus lacustris (L.) Palla (*Scirpus lacustris* L.) , Common Club-rush
Sparganium emersum Rehmann, Unbranched Bur-reed
Sparganium natans L. (*S. minimum* Wallr.), Least Bur-reed
Subularia aquatica L., Awlwort
Typha latifolia L., Bulrush
Utricularia intermedia Hayne, Intermediate Bladderwort
Utricularia vulgaris L., Greater Bladderwort

Appendix C: Abiotic Characterisitcs of the Study Sites

Table C.1 Sediment composition

	% Composition by weight					
Sample	Clay	Silt	Fine sand	Coarse sand	Gravel	% organic matter
Dee 1 a	13.93	11.01	29.19	45.16	3.40	3.31
Dee 1 b	14.98	8.73	27.12	46.58	3.40	3.42
Dee 2 a	13.57	8.40	40.86	32.88	6.20	5.52
Dee 2 b	14.75	8.78	37.99	34.98	6.20	6.00
Dee 3 a	6.51	3.81	39.08	39.09	12.90	0.54
Dee 3 b	6.21	3.49	38.42	40.33	12.90	0.54
Lom 1 a	7.40	1.67	48.75	42.77	1.80	8.14
Lom 1 b	7.92	1.80	49.72	40.02	1.80	8.15
Lom 2 a	32.40	31.52	28.40	2.56	0.00	10.36
Lom 2 b	33.57	30.64	26.76	2.45	0.00	10.01
Lom 3 a	35.80	32.48	23.23	1.50	0.00	10.78
Lom 3 b	35.50	33.42	22.97	1.58	0.00	10.78
Men 1 a	2.79	0.47	16.08	15.09	67.60	2.15
Men 1 b	2.75	0.67	14.71	16.54	67.60	2.04
Men 2 a	8.36	1.97	73.13	19.07	3.80	0.86
Men 2 b	8.03	1.76	70.93	21.26	3.80	0.87
Men 3 a	8.08	2.98	20.42	46.00	25.30	3.94
Men 3 b	8.06	1.65	17.81	48.54	25.30	4.06
Low 1 a	7.38	1.00	72.86	26.30	1.80	1.02
Low 1 b	7.47	0.85	75.07	21.43	1.80	0.98
Low 2 a	9.38	3.47	82.98	10.72	0.10	0.70
Low 2 b	8.57	3.64	86.29	7.15	0.10	0.68
Low 3 a	8.44	2.87	87.20	8.34	0.10	1.06
Low 3 b	8.55	2.71	84.65	11.47	0.10	1.12

Table C.2 Siltation Rate

Plot	Dates	silt deposited g/m2/day			mean	st err
Dee 1	15/05/90 to 30/10/90	0.25	0.62	0.27	0.38	0.12
	30/10/90 to 21/03/91		0.21	0.28	0.24	0.03
	21/03/91 to 13/08/91		0.23	0.25	0.24	0.01
Dee2	15/05/90 to 30/10/90	0.22	0.19	0.15	0.19	0.02
	30/10/90 to 21/03/91	0.56	0.19	0.21	0.32	0.12
	21/03/91 to 13/08/91	0.21	0.18	0.25	0.21	0.02
Dee3	15/05/90 to 30/10/90	0.07	0.24	0.16	0.16	0.05
	30/10/90 to 21/03/91	0.38	0.24	0.44	0.35	0.06
	21/03/91 to 13/08/91	0.22	0.22	0.22	0.22	0.00
Lom 1	26/06/90 to 09/11/90	0.47	0.68	0.99	0.71	0.15
	09/11/90 to 19/03/91	0.19	0.31	0.81	0.44	0.19
	19/03/91 to 20/08/91	0.26	0.81	1.17	0.74	0.27
Lom 2	26/06/90 to 09/11/90	0.28	0.29		0.29	0.01
	09/11/90 to 19/03/91	0.26	0.23		0.24	0.02
	19/03/91 to 20/08/91	0.12	0.15		0.14	0.02
Lom 3	26/06/90 to 19/03/91	0.58	0.13		0.36	0.23
	19/03/91 to 20/08/91	0.44	0.38		0.41	0.03
Men 1	23/05/90 to 14/11/90	0.27	0.53	0.56	0.45	0.09
	14/11/90 to 03/04/91	0.48	0.63	0.59	0.57	0.04
	03/04/91 to 27/08/91	0.60	0.76	0.59	0.65	0.05
Men 2	14/11/90 to 03/04/91	0.51	0.55	0.47	0.51	0.02
	03/04/91 to 27/08/91	0.33	0.32	0.31	0.32	0.01
Men 3	23/05/90 to 14/11/90	0.04	0.06	0.23	0.11	0.06
	14/11/90 to 03/04/91	0.10	0.18	0.25	0.18	0.04
Low 1	10/07/90 to 23/11/90	2.41	2.77	5.42	3.53	0.95
	23/11/90 to 04/04/91	2.90	2.05	3.01	2.65	0.30
	04/04/91 to 03/10/91	0.86	1.28	1.44	1.19	0.17
Low 2	29/05/90 to 23/11/90	0.53	1.28		0.90	0.38
	23/11/90 to 04/04/91	0.87	0.96		0.91	0.05
	04/04/91 to 03/10/91	0.36	0.58		0.47	0.11
Low 3	29/05/90 to 23/11/90	1.95	0.73	0.99	1.22	0.37
	23/11/90 to 04/04/91	1.13	1.39	0.90	1.14	0.14
	04/04/91 to 03/10/91	0.51	0.39	0.45	0.45	0.03

Table C.3 Water Physico-chemistry

Dee Plot 1

Date	pH	Cond. µs/cm	E	Temp (°C)	Dissolved Oxygen mg/l	% Sat
4/4/90	6.3	53.1	1.39	6	9.0	59
15/5/90	5.1	56.0		14	12.6	
18/6/90	5.3		0.78	15	12.4	120
7/8/90	5.5	42.9		14	11.2	110
2/10/90	6.0			9	9.8	89
9/4/91	5.1	39.0	0.90	6	12.4	103
11/6/91	5.7	35.6	1.24	10	9.7	84
9/7/91	5.7	34.6	1.17	13		
13/8/91		31.0	0.93	14		84

Dee Plot 2

Date	pH	Cond. µs/cm	E	Temp (°C)	Dissolved Oxygen mg/l	% Sat
15/5/90	5.1	50.3		14	12.9	129
18/6/90	5.4		1.12	15	12.4	116
7/8/90	5.6	42.6		14	10.9	104
9/4/91	5.1	39.0	0.90	6	12.4	103
4/6/91	5.7	39.2	1.09	11	10.6	97
9/7/91	5.7	34.6	1.20	13		
13/8/91		31.0	1.02	14		84

Dee Plot 3

Date	pH	Cond. µs/cm	E	Temp (°C)	Dissolved Oxygen mg/l	% Sat
15/5/90	5.1	53.4		14	12.4	118
18/6/90	5.3		2.20	14	13.4	129
7/9/90	5.6	42.8		16	10.7	117
9/4/91	5.5	39.0	0.83	6	13.6	106
4/6/91	5.7	39.4	1.04	11	10.5	98
9/7/91	5.9	34.6	1.01	15		
13/8/91		32.6	0.85	15		86

Lomond Plot 1

Date	pH	Cond. µs/cm	E	Temp (°C)	Dissolved Oxygen mg/l	% Sat
6/4/90	7.8	51.0	1.13	7	6.9	56
8/5/90	6.9		0.81	12	5.7	
26/6/90	7.1	38.0	0.97	14	10.2	106
23/7/90	6.5	60.0	0.11	19	8.6	95
25/9/90	6.9	110.0	1.23	13	5.7	53
17/4/91	6.6	59.1	0.98	6	13.2	102
10/6/91	6.2	54.7	0.90	12	10.5	100
18/7/91		52.9	1.09	15		98
20/8/91	6.8	52.9	1.10	16		86

Lomond Plot 2

Date	pH	Cond. µs/cm	E	Temp (°C)	Dissolved Oxygen mg/l	% Sat
8/5/90	6.5		0.91	12	5.1	
26/6/90	7.2	35.0	0.98	14	10.6	107
23/7/90	6.5	59.0	0.21	19	8.4	92
25/9/90	6.9	40.0	1.14	13	4.4	43
17/4/91	6.3	58.3	1.03	6	10.4	103
10/6/91	6.2	55.9	0.86	13	10.7	96
18/7/91		52.7	1.09	15		98
20/8/91	6.8	52.4	0.98	16		84

Lomond Plot3

Date	pH	Cond. µs/cm	E	Temp (°C)	Dissolved Oxygen mg/l	% Sat
8/5/90	7.2		1.14	12	4.2	
26/6/90	7.3	35.0	0.75	14	10.5	104
23/7/90	6.7	60.0	0.38	20	8.4	91
25/9/90	7.1	40.0	2.08	13	5.5	52
17/4/91	6.4	59.0	1.24	6	10.5	105
10/6/91	6.2	55.8	0.83	13	10.4	100
18/7/91		53.2	0.98	15		98
20/8/91	6.8	53.0	0.78	16		85

Menteith Plot 1

Date	pH	Cond. µs/cm	E	Temp (°C)	Dissolved Oxygen mg/l	% Sat
19/4/90	6.8	77.6	1.85	7	7.4	55
23/5/90	8.1	74.8	1.28	13		
3/7/90	6.9	61.0	2.34	15	10.2	98
28/8/90	6.8	79.5	0.18	19		
10/10/90	6.6	78.9	3.00	10	10.8	93.8
1/5/91	7.7	77.4	1.41	9	13.1	111
18/6/91	7.7	82.1	0.52	14	11.4	112
23/7/91	7.0	82.2	1.25	17		104
27/8/91	6.8	84.1	1.05	17		83

Menteith Plot 2

Date	pH	Cond. µs/cm	E	Temp (°C)	Dissolved Oxygen mg/l	% Sat
3/7/90	6.8	60.0	2.39	15	13.2	130
28/8/90	6.9	78.0	0.21	19	6.6	152 III
10/10/90	6.5	79.3	0.95	10	10.5	95
1/5/91	7.6	77.4	1.47	9	13.5	118
18/6/91	7.8	93.1	0.18	14	12.1	119
23/7/91	7.0	80.6	1.61	17		104
27/8/91	6.8	84.1	1.06	17		83

Menteith Plot 3

Date	pH	Cond. µs/cm	E	Temp (°C)	Dissolved Oxygen mg/l	% Sat
19/4/90	6.9	65.3	1.30	7	7.4	58
23/5/90	7.4	73.5	0.95	13		
3/7/90	6.9	60.0	1.50	15	13.8	127
28/8/90	7.5	77.7	0.24	19		
10/10/90	6.5	78.9	2.65	9	13.1	115
1/5/91	7.8	77.8	1.43	10	13.1	116
18/6/91	7.8	83.0	0.14	13	12.4	120
23/7/91	7.0	82.2	1.45	17		104
27/8/91	6.8	84.7	0.99	17		84

Lowes Plot 1

Date	pH	Cond. µs/cm	E	Temp (°C)	Dissolved Oxygen mg/l	% Sat
18/4/90	6.6	85.0	1.47	8	8.3	51
29/5/90	8.2	84.2	0.69	15		
10/7/90	6.7		0.10	14	9.9	118
4/9/90	6.6	89.0	0.10	17	7.5	77
15/10/90	6.9	92.1	1.30	12	8.8	62
23/4/91	6.9		0.70	9	12.2	107
25/6/91	9.1	95.2	1.20	15	11.2	109
30/7/91	6.6	94.0	1.33	18		84
3/9/91	7.2	94.3	1.33	18		94

Lowes Plot 2

Date	pH	Cond. µs/cm	E	Temp (°C)	Dissolved Oxygen mg/l	% Sat
29/5/90	8.5	84.1	0.51	15		
10/7/90	6.8		0.12	15		
4/9/90	6.8	88.4	0.09	17	9.2	93
15/10/90	6.7	90.8	0.85	10	7.8	71
23/4/91	6.9		1.07	9	13.2	110
25/6/91	8.9	96.4	1.18	14	12.3	114
30/7/91	6.5	94.6	1.54	19		84
3/9/91	7.6	93.6	1.12	18		92

Lowes Plot 3

Date	pH	Cond. µs/cm	E	Temp (°C)	Dissolved Oxygen mg/l	% Sat
18/4/90			1.01			
29/5/90	8.1	84.7	0.64	15		
10/7/90	6.9		0.20	15		
4/9/90	6.8	88.3	0.13	17	5.5	56
15/10/90	7.8	90.7	2.00	10	7.8	
23/4/90	6.9		1.46	8	13.0	105
25/6/91	8.7	95.7	0.99	14	11.8	119
30/7/91	6.5	94.0	1.08	18		84
3/9/91	7.7	93.6	0.94	18		100

APPENDIX D:

RAW FIELD DATA

Abbreviations used in Table D.1 *Littorella* Biomass

Date	Date of sample collection
Wt 5	Oven dry weight of 5 intact plants
Tot. samp	Dry weight of entire sample expressed in gm^{-2}
% Litt	Contribution of *Littorella* to biomass expressed as % oven dry weight of sample
R:S	Ratio between roots & shoots expressed on a dry weight basis
S Err	Standard Error of Mean

Table D.1 *Littorella* Biomass

Dee Plot1 1990

Date	Wt 5	Tot. samp	% Litt	R:S	Date	Wt 5	Tot. samp	% *Litt*	R:S
16-05		8.8	85.0		18-06		26.0	88.1	0.50
		16.7	100.0	0.50			33.4	84.2	1.00
		4.8	18.2				57.2	100.0	1.00
		44.4	79.2	1.11			49.7	91.2	
Mean		18.7	70.6	0.81	Mean		41.6	90.1	0.83
S Err		7.7	15.6		S Err		6.2	2.9	
Date	Wt 5	Tot. Samp	% Litt	R:S	Date	Wt 5	Tot. samp	% *Litt*	R:S
07-08		1.1	75.0	1.29	02-10				
		1.1	56.4	1.00					
		1.7	00.6						
Mean		21.1	44.0	1.14	Mean				
S Err		2.3	18.2		S Err				

Dee Plot1 1991

Date	Wt 5	Tot. Samp	% Litt	R:S	Date	Wt 5	Tot. samp	% *Litt*	R:S
09-04		22.2	94.2	0.67	04-06	0.19	15.8	57.6	1.00
		47.8	28.8	0.71		0.08	66.4	74.0	
		54.7	83.0	1.37		0.16	47.8	86.6	0.53
		50.2	97.4	1.26		0.19	57.1	96.9	
Mean		43.8	75.9	1.00	Mean	0.15	46.8	78.8	0.76
S Err		6.3	13.9		S Err	0.02	9.5	7.3	
Date	Wt 5	Tot. Samp	% Litt	R:S	Date	Wt 5	Tot. samp	% *Litt*	R:S
09-07	0.26	48.3	90.7	1.06	13-08	0.10	44.0	45.1	0.70
	0.39	54.9	98.5	1.03		0.09	40.6	66.1	0.78
	0.90	70.4	83.0	1.28		0.11	38.4	60.8	0.60
	0.14	40.8	95.7	1.48		0.13	30.7	60.4	0.69
Mean	0.42	53.6	92.0	1.21	Mean	0.11	38.4	58.1	0.69
S Err	0.14	5.4	3.0		S Err	0.01	2.4	3.9	

Dee Plot2 1990

Date	Wt 5	Tot. Samp	% Litt	R:S
16-05		38.7	53.4	0.63
		158.8	100.0	0.86
		32.6	97.3	0.83
		18.9	74.4	
Mean		62.3	81.3	0.77
St Err		28.1	9.5	

Date	Wt 5	Tot. samp	% *Litt*	R:S
18-06		189.6	94.4	1.48
		40.9	100.0	0.86
		194.4	96.8	1.5
		208.6	94.7	
Mean		158.3	96.5	1.28
St Err		34.1	1.1	

Date	Wt 5	Tot. Samp	% Litt	R:S
07-08	0.12	59.7	74.8	1.33
	0.14	109.8	93.3	1.00
	0.20	106.9	94.5	1.22
Mean	0.15	92.1	87.5	1.19
St Err	0.02	13.3	5.2	

Date	Wt 5	Tot. samp	% *Litt*	R:S
02-10				
Mean				
St Err				

Dee Plot2 1991

Date	Wt 5	Tot. Samp	% Litt	R:S
09-04				
Mean				
St Err				

Date	Wt 5	Tot. samp	% *Litt*	R:S
04-06	0.13	78.1	94.5	1.97
	0.14	37.6	100.0	1.22
	0.17	60.1	70.5	1.44
Mean	0.15	58.8	88.3	1.54
St Err	0.01	9.6	7.4	0.18

Date	Wt 5	Tot. Samp	% Litt	R:S
09-07	0.14	35.8	42.9	0.61
	0.13	19.2	50.8	0.66
	0.12	39.5	83.0	0.53
	0.14	60.2	68.6	0.69
Mean	0.13	38.7	61.3	0.62
St Err	0.00	7.3	7.8	

Date	Wt 5	Tot. samp	% *Litt*	R:S
13-08	0.18	58.3	88.5	0.74
	0.11	49.6	95.8	0.72
	0.14	111.4	100.0	0.92
	0.19	62.6	93.6	0.41
Mean	0.15	70.4	94.5	0.70
St Err	0.02	12.0	2.1	

Dee Plot3 1990

Date	Wt 5	Tot. Samp	% Litt	R:S
16-05		18.5	28.6	
		10.6	25.0	
		11.0	68.0	1.00
		12.3	35.7	
Mean		13.1	39.3	1.00
St Err		1.6	8.5	

Date	Wt 5	Tot. samp	% *Litt*	R:S
18-06		22.9	21.2	1.00
		26.4	20.0	0.60
		20.7	38.3	1.00
		17.6	17.5	
Mean		21.9	12.2	0.87
St Err		1.6	4.1	

Date	Wt 5	Tot. Samp	% Litt	R:S
07-08	0.08	41.1	61.1	1.00
	0.04	49.8	79.7	1.00
	0.05	29.9	36.4	1.50
Mean	0.06	40.3	59.1	1.17
St Err	0.01	4.7	10.3	

Date	Wt 5	Tot. samp	% *Litt*	R:S
02-10	0.04	29.1	6.0	1.00
	0.02	29.8	61.3	1.00
	0.02	15.8	14.1	1.00
	0.05	14.2	32.6	1.50
Mean	0.03	22.2	28.5	1.13
St Err	0.01	3.6	10.6	

Dee Plot3 1991

Date	Wt 5	Tot. Samp	% Litt	R:S
09-04		51.8	26.5	0.83
		35.0	7.8	0.89
		18.1	36.3	1.05
		55.2	11.0	0.90
Mean		40.0	20.4	0.92
St Err		7.4	5.8	0.04

Date	Wt 5	Tot. samp	% *Litt*	R:S
04-06	0.06	27.7	48.0	.050
	0.06	26.2	43.9	1.08
	0.03	21.9	11.7	1.17
	0.04	26.7	19.2	0.87
Mean	0.05	25.6	30.7	0.90
St Err	0.01	1.1	7.8	0.13

Date	Wt 5	Tot. Samp	% Litt	R:S
09-07	0.06	18.9	61.9	0.86
	0.06	12.2	63.2	0.91
	0.05	13.0	17.3	
	0.07	20.5	21.9	1.10
Mean	0.06	16.1	41.0	0.96
St Err	0.00	1.8	10.8	

Date	Wt 5	Tot. samp	% *Litt*	R:S
13-08	0.06	12.2	39.5	0.71
	0.04	9.6	50.0	0.88
	0.04	20.5	64.1	1.06
	0.05	11.8	58.1	1.45
Mean	0.05	13.5	52.9	1.03
St Err	0.00	2.1	4.6	

Lomond Plot1 1990

Date	Wt 5	Tot. Samp	% Litt	R:S
08-05		72.6	100.0	1.22
		133.3	100.0	1.23
		108.2	100.0	1.71
Mean		104.7	100.0	1.39
St Err		14.4	0.0	

Date	Wt 5	Tot. Samp	% Litt	R:S
26-06		96.8	86.4	1.11
		40.0	63.7	1.25
		45.3	47.6	
		84.9	99.5	1.08
Mean		66.8	74.3	0.07
St Err		12.3	10.0	

Date	Wt 5	Tot. Samp	% Litt	R:S
22-07		94.2	96.3	0.93
		99.4	86.6	0.89
		189.4	92.3	0.86
Mean		127.7	91.7	0.89
St Err		25.2	2.3	

Date	Wt 5	Tot. Samp	% Litt	R:S
25-09		87.4	85.5	1.88
		207.2	99.9	0.47
		80.0	100.0	1.05
		198.1	98.9	1.33
Mean		142.2	96.1	1.18
St Err		29.4	3.1	

Lomond Plot1 1991

Date	Wt 5	Tot. Samp	% Litt	R:S
17-04		46.7	100.0	0.92
	0.17	68.5	100.0	1.42
	0.18	33.0	100.0	0.76
	0.17	82.4	77.5	0.81
Mean	0.17	57.6	94.4	0.98
St Err	0.00	9.6	4.9	

Date	Wt 5	Tot. Samp	% Litt	R:S
10-06	0.17	83.8	100.0	1.05
	0.16	55.4	99.1	0.84
	0.18	161.8	100.0	1.19
	0.12	145.8	100.0	1.07
Mean	0.16	111.7	99.8	1.04
St Err	0.01	21.8	0.2	

Date	Wt 5	Tot. Samp	% Litt	R:S
18-07	0.18	215.8	100.0	1.04
		195.5		
	0.24	126.2	94.9	1.25
	0.28	322.6		
Mean	0.23	215.0	97.5	1.15
St Err	0.02	35.2	1.8	0.07

Date	Wt 5	Tot. Samp	% Litt	R:S
20-08	0.36	116.6	100.0	0.90
	0.18	104.6	97.4	0.94
	0.28	201.6	98.2	0.76
	0.21	163.0	99.1	0.78
Mean	0.26	146.5	98.7	0.84
St Err	0.03	19.3	0.5	

Lomond Plot2 1990

Date	Wt 5	Tot. Samp	% Litt	R:S
08-05		98.6	64.7	1.36
		150.0	78.3	1.43
		146.5	65.5	
		41.8	94.7	2.50
Mean		109.2	75.8	1.76
St Err		22.0	6.1	

Date	Wt 5	Tot. Samp	% Litt	R:S
26-06		114.8	69.7	0.90
		72.2	78.7	1.25
		147.0	51.5	1.20
Mean		111.3	66.6	1.12
St Err		17.7	6.5	

Date	Wt 5	Tot. Samp	% Litt	R:S
22-07		95.5	65.3	1.25
		40.5	31.6	1.75
		157.4	95.8	1.67
Mean		97.8	64.3	1.56
St Err		27.3	15.1	

Date	Wt 5	Tot. Samp	% Litt	R:S
25-09		141.3	99.1	0.76
		159.2	93.5	1.50
		174.1	100.0	1.60
		98.7	95.0	1.44
Mean		143.3	96.9	1.33
St Err		14.1	1.4	

Lomond Plot2 1991

Date	Wt 5	Tot. Samp	% Litt	R:S
17-04		202.2	100.0	1.02
	0.15	204.3	100.0	0.95
	0.18	191.0	100.0	0.87
	0.18	144.6	100.0	0.90
Mean	0.17	185.6	100.0	0.93
St Err	0.01	12.1	0.0	

Date	Wt 5	Tot. Samp	% Litt	R:S
10-06	0.18	72.3	94.9	1.30
	0.12	57.8	79.2	1.38
	0.16	56.5	94.0	1.28
	0.11	111.4	97.6	1.33
Mean	0.14	74.5	91.4	1.32
St Err	0.01	11.1	3.6	

Date	Wt 5	Tot. Samp	% Litt	R:S
18-07	0.23	143.2	99.8	1.19
	0.23	70.2		1.15
	0.19	116.2	97.5	
	0.10	32.2		
Mean	0.19	90.4	98.6	1.17
St Err	0.03	21.3	0.8	0.02

Date	Wt 5	Tot. Samp	% Litt	R:S
20-08	0.28	133.6	99.8	1.63
	0.18	135.5	97.4	0.94
	0.19	61.0	87.4	1.15
	0.29	139.8	95.3	1.07
Mean	0.23	117.5	94.9	1.20
St Err	0.03	16.4	2.3	0.13

Lomond Plot3 1990

Date	Wt 5	Tot. Samp	% Litt	R:S
18-05		117.0	45.9	1.40
		105.2	69.9	2.20
		106.0	90.0	1.00
Mean		109.4	68.6	1.53
St Err		3.1	10.4	

Date	Wt 5	Tot. Samp	% Litt	R:S
26-06		79.2	4.4	
		114.8	86.2	1.35
		59.4	96.3	0.93
		59.4	96.3	0.93
Mean		78.2	70.8	1.07
St Err		11.3	19.3	

Date	Wt 5	Tot. Samp	% Litt	R:S
22-07		127.5	66.9	1.29
		114.7	58.3	1.50
		81.6	62.2	1.29
Mean		108.0	62.4	1.36
St Err		11.2	2.0	

Date	Wt 5	Tot. Samp	% Litt	R:S
25-09		132.0	98.4	1.5
		196.8	100.0	1.2
		133.9	98.2	1.2
		141.1	100.0	
Mean		151.0	99.2	1.3
St Err		13.3	0.4	

Lomond Plot3 1991

Date	Wt 5	Tot. Samp	% Litt	R:S
17-04		152.0	100.0	0.88
	0.16	154.9	100.0	0.79
	0.17	170.2	100.0	0.86
	0.24	141.3	97.1	0.76
Mean	0.19	154.6	99.3	0.82
St Err	0.02	5.2	0.6	

Date	Wt 5	Tot. Samp	% Litt	R:S
10-06	0.13	134.4	100.0	0.85
	0.16	78.6	94.7	
	0.12	176.6	100.0	1.23
	0.21	171.4	100.0	1.06
Mean	0.15	140.2	98.7	1.05
St Err	0.02	19.6	1.2	

Date	Wt 5	Tot. Samp	% Litt	R:S
18-07	0.12	132.2		
	0.17	114.6	96.8	1.11
	0.11	107.4	94.2	1.85
	0.14	89.6	100.0	1.17
Mean	0.13	110.9	97.0	1.38
St Err	0.01	7.6	1.4	0.19

Date	Wt 5	Tot. Samp	% Litt	R:S
20-08	0.21	137.6	92.1	1.03
	0.17	130.4	90.4	0.99
	0.27	133.6	95.8	1.43
	0.21	141.8	85.7	1.49
Mean	0.21	135.8	91.0	1.23
St Err	0.02	2.1	1.8	

Menteith Plot1 1990

Date	Wt 5	Tot. Samp	% Litt	R:S
23-05		41.4	63.8	0.67
		34.3	76.9	0.75
		44.0	79.0	
		38.7	52.3	
Mean		39.6	67.7	0.71
St Err		1.8	5.4	

Date	Wt 5	Tot. Samp	% Litt	R:S
03-07	0.11	40.9	59.1	0.83
	0.14	40.5	83.7	0.75
	0.12	33.0	68.0	1.40
Mean	0.12	38.1	70.3	0.99
St Err	0.01	2.1	5.9	

Date	Wt 5	Tot. Samp	% Litt	R:S
28-08		64.2	85.8	1.33
		57.9	51.9	1.18
		37.4	79.9	1.00
		73.0	55.3	1.00
Mean		58.1	68.2	1.13
St Err		6.5	7.4	

Date	Wt 5	Tot. Samp	% Litt	R:S
10-10	0.11	73.8	80.5	1.36
		86.9	79.6	1.23
	0.11	86.2	91.8	1.45
	0.15	77.3	92.3	1.38
Mean	0.12	81.0	86.0	1.35
St Err	0.01	2.8	3.0	

Menteith Plot1 1991

Date	Wt 5	Tot. Samp	% Litt	R:S
01-05	0.09	84.8	12.1	0.49
	0.14	58.2	42.3	0.70
	0.10	80.5	6.4	0.57
	0.10	66.9	37.3	0.59
Mean	0.11	72.6	24.5	0.59
St Err	0.01	5.3	7.8	

Date	Wt 5	Tot. Samp	% Litt	R:S
18-06	0.09	33.8	38.9	1.14
	0.09	65.4	15.9	0.85
	0.09	53.0	31.4	1.23
	0.09	107.4	5.4	0.93
Mean	0.09	64.9	22.9	1.04
St Err	0.00	13.5	6.5	

Date	Wt 5	Tot. Samp	% Litt	R:S
23-07	0.07	124.0	14.7	1.28
	0.11	149.4	5.5	0.82
	0.06	39.0	85.2	0.80
	0.07	57.0	57.6	0.80
Mean	0.08	92.4	40.8	0.92
St Err	0.01	22.8	16.2	

Date	Wt 5	Tot. Samp	% Litt	R:S
27-08	0.06	48.3	45.4	1.30
	0.12	76.6	21.1	0.89
	0.11	38.9	93.8	0.94
	0.08	45.3	64.3	1.20
Mean	0.09	52.3	56.2	1.08
St Err	0.01	7.2	13.3	0.09

Menteith Plot2 1990

Date	Wt 5	Tot. Samp	% Litt	R:S
23-05				
Mean				
St Err				

Date	Wt 5	Tot. Samp	% Litt	R:S
03-07	0.13	70.8	33.5	1.17
		19.4	52.3	
	0.04	30.4	29.0	1.00
	0.12	51.0	31.0	1.40
Mean	0.10	42.9	36.5	1.19
St Err	0.02	9.9	4.6	

Date	Wt 5	Tot. Samp	% Litt	R:S
28-08		49.4	31.1	1.50
		75.5	40.0	
		40.2	39.4	1.00
		79.4	55.8	1.50
Mean		61.1	41.6	1.33
St Err		8.4	4.5	

Date	Wt 5	Tot. Samp	% Litt	R:S
10-10		65.9	88.8	1.62
		148.0	96.3	
	0.09	82.6	89.2	1.50
	0.11	105.6	91.2	1.53
Mean	0.10	100.5	91.4	1.55
St Err	0.01	15.4	1.5	

Menteith Plot2 1991

Date	Wt 5	Tot. Samp	% Litt	R:S
01-05	0.08	58.4	72.0	1.24
	0.09	65.6	54.2	1.70
	0.06	82.1	78.0	1.36
	0.09	58.4	71.2	1.51
Mean	0.08	66.1	68.8	1.45
St Err	0.01	4.8	4.4	

Date	Wt 5	Tot. Samp	% Litt	R:S
18-06		28.2	93.2	
		25.3	98.7	
	0.11	35.8	92.0	
	0.08	41.0	63.3	
Mean	0.09	32.6	86.8	
St Err	0.01	3.1	6.9	

Date	Wt 5	Tot. Samp	% Litt	R:S
23-07	0.14	55.5	88.2	1.12
	0.11	48.5	83.5	1.27
	0.13	43.2	90.7	1.16
	0.15	67.4	93.8	1.80
Mean	0.13	53.6	89.1	1.34
St Err	0.01	4.5	1.9	

Date	Wt 5	Tot. Samp	% Litt	R:S
17-08	0.15	45.9	70.4	0.93
	0.14	45.8	79.0	0.96
	0.09	121.4	38.2	1.05
	0.09	28.6	82.1	0.89
Mean	0.12	60.4	67.4	0.96
St Err	0.01	18.0	8.7	0.03

Menteith Plot3 1990

Date	Wt 5	Tot. Samp	% Litt	R:S
23-05		55.9	10.2	0.33
		52.8	17.5	1.00
		125.8	25.5	1.33
Mean		78.2	17.8	0.89
St Err		19.5	3.6	

Date	Wt 5	Tot. Samp	% Litt	R:S
03-07	0.05	47.5	35.2	0.67
		53.7	8.2	
	0.04	29.5	14.9	1.00
	0.08	38.7	18.2	0.60
Mean	0.06	42.4	19.1	0.76
St Err	0.01	4.6	5.0	

Date	Wt 5	Tot. Samp	% Litt	R:S
28-08		74.2	6.0	1.67
		243.8	3.5	0.67
		724.8	5.6	1.00
		514.9	11.8	1.00
Mean		389.4	6.7	1.08
St Err		124.7	1.5	

Date	Wt 5	Tot. Samp	% Litt	R:S
10-10	0.11	298.6	2.2	1.00
		394.9	12.0	1.00
		402.2	18.2	
		428.6	8.7	0.81
Mean	0.11	381.1	10.3	0.94
St Err	0.00	24.6	2.9	

Menteith Plot3 1991

Date	Wt 5	Tot. Samp	% Litt	R:S
01-05	0.04	69.0	13.5	1.08
	0.07	98.1	9.3	1.00
	0.04	91.4	2.3	0.50
	0.06	90.1	9.1	0.88
Mean	0.05	87.1	8.5	0.86
St Err	0.01	5.5	2.0	

Date	Wt 5	Tot. Samp	% Litt	R:S
18-06	0.12	122.7	36.0	
	0.11	150.2	25.6	
	0.15	116.2	30.0	
		176.6	23.7	1.04
Mean	0.13	141.4	28.8	
St Err	0.01	12.0	2.4	1.04

Date	Wt 5	Tot. Samp	% Litt	R:S
23-07	0.15	114.1	27.5	1.20
	0.08	99.5	22.8	1.39
	0.18	114.2	24.0	
Mean	0.14	109.2	24.8	1.29
St Err	0.02	4.0	1.2	

Date	Wt 5	Tot. Samp	% Litt	R:S
27-08	0.10	188.6	14.7	0.87
	0.24	236.6	28.1	0.75
	0.19	292.2	10.2	0.64
	0.17	333.9	15.0	0.67
Mean	0.17	262.8	17.0	0.73
St Err	0.03	27.5	3.4	

Lowes Plot1 1990

Date	Wt 5	Tot. Samp	% Litt	R:S
29-05	0.02	20.7	83.0	1.00
	0.04	39.6	20.0	1.00
	0.02	33.4	61.8	1.00
	0.03	35.2	47.5	0.50
Mean	0.03	32.2	53.1	0.88
St Err	0.00	3.5	11.4	

Date	Wt 5	Tot. Samp	% Litt	R:S
10-07	0.04	18.9	37.2	1.00
Mean	0.04	18.9	37.2	1.00
St Err				

Date	Wt 5	Tot. Samp	% Litt	R:S
04-09		70.6	76.2	0.77
		33.0	74.8	
		45.9	49.1	1.33
		64.0	38.8	2.00
Mean		53.4	59.7	1.37
St Err		7.4	8.1	

Date	Wt 5	Tot. Samp	% Litt	R:S
15-10		65.0	43.6	1.13
		63.8	56.4	1.60
		89.1	32.3	2.44
		65.6	39.3	1.57
Mean		70.9	42.9	1.69
St Err		5.3	4.4	

Lowes Plot1 1991

Date	Wt 5	Tot. Samp	% Litt	R:S
23-04	0.12	24.2	62.9	0.89
	0.15	26.2	40.9	0.93
	0.11	59.2	38.4	0.82
	0.11	45.4	40.1	0.75
Mean	0.12	38.8	45.6	0.84
St Err	0.01	7.2	5.0	

Date	Wt 5	Tot. Samp	% Litt	R:S
25-06	0.09	21.9	39.4	1.25
	0.08	48.3	98.2	1.32
	0.08	21.9	67.9	1.30
	0.06	25.0	79.5	1.15
Mean	0.08	29.3	63.8	1.25
St Err	0.01	5.5	7.4	

Date	Wt 5	Tot. Samp	% Litt	R:S
30-07	0.17	38.9	42.0	1.03
	0.12	40.0	44.0	1.05
	0.13	14.6	79.1	0.79
	0.07	17.4	72.5	0.85
Mean	0.12	27.7	59.4	0.93
St Err	0.02	5.9	8.3	

Date	Wt 5	Tot. Samp	% Litt	R:S
03-09	0.09	44.3	68.6	0.87
	0.09	17.1	33.6	1.08
	0.11	23.0	66.7	1.24
	0.07	9.4	71.2	0.94
Mean	0.09	23.5	60.0	1.03
St Err	0.01	6.5	7.7	

Lowes Plot2 1990

Date	Wt 5	Tot. Samp	% Litt	R:S
29-05	0.04	23.8	44.4	3.00
	0.05	42.2	77.1	1.50
	0.02	78.5	30.5	0.25
		50.2	39.5	
Mean	0.04	48.7	47.9	1.58
St Err	0.01	9.9	8.8	

Date	Wt 5	Tot. Samp	% Litt	R:S
10-07	0.05	27.2	45.2	1.50
	0.06	23.3	35.9	1.00
	0.05	26.4	56.7	0.67
		10.6	41.7	
Mean		21.9	44.8	1.06
St Err		3.4	3.8	

Date	Wt 5	Tot. Samp	% Litt	R:S
04-09		65.3	63.0	1.00
		51.7	41.2	1.50
		77.4	73.1	1.50
		35.4	48.0	1.25
Mean		57.4	56.3	1.31
St Err		7.8	6.3	

Date	Wt 5	Tot. Samp	% Litt	R:S
15-10		49.3	59.1	0.83
		52.2	36.2	1.5
		53.3	41.1	
		68.3	40.8	
Mean		55.8	44.3	1.17
St Err		3.7	4.4	

Lowes Plot2 1991

Date	Wt 5	Tot. Samp	% Litt	R:S
23-04	0.15	67.8	57.3	1.30
	0.11	50.1	69.0	1.02
	0.08	58.2	60.2	0.94
	0.11	58.4	62.5	1.05
Mean	0.11	58.6	62.2	1.08
St Err	0.01	3.1	2.2	

Date	Wt 5	Tot. Samp	% Litt	R:S
25-06	0.09	24.3	33.6	1.00
	0.07	11.2	65.7	1.05
	0.09	24.4	50.3	0.84
	0.08	21.9	32.8	0.95
Mean	0.08	20.5	45.6	0.96
St Err	0.00	2.7	6.8	

Date	Wt 5	Tot. Samp	% Litt	R:S
30-07	0.07	12.8	91.2	1.45
	0.11	38.9	96.3	1.42
	0.08	25.4	88.0	1.57
	0.09	21.3	81.2	1.35
Mean	0.09	24.6	89.2	1.45
St Err	0.01	4.7	2.7	

Date	Wt 5	Tot. Samp	% Litt	R:S
03-09	0.08	8.2	80.4	1.06
	0.07	22.1	60.1	1.05
	0.06	17.6	53.6	0.96
	0.04	19.0	23.5	1.00
Mean	0.06	16.7	54.4	1.02
St Err	0.01	2.6	10.2	

Lowes Plot3 1990

Date	Wt 5	Tot. Samp	% Litt	R:S
29-05		19.4	13.6	
	0.05	83.6	30.0	0.67
	0.06	106.5	17.4	0.50
	0.05	40.5	19.6	0.67
Mean	0.05	62.5	20.1	0.61
St Err	0.00		3.0	

Date	Wt 5	Tot. Samp	% Litt	R:S
10-07	0.07	13.6	74.2	0.75
		10.1	56.5	
	0.05	4.8	90.9	1.50
	0.08	28.2	57.8	1.00
Mean	0.07	14.2	69.9	1.08
St Err	0.01	4.3	7.0	

Date	Wt 5	Tot. Samp	% Litt	R:S
04-09	0.07	56.2	16.5	0.75
	0.12	69.4	31.6	0.71
	0.08	85.4	25.7	0.60
	0.14	90.1	26.1	0.75
Mean	0.10	75.3	25.0	0.70
St Err	0.01	6.7	2.7	

Date	Wt 5	Tot. Samp	% Litt	R:S
15-10		45.9	44.2	0.83
		53.3	42.6	1.22
		59.5	48.7	
		58.9	42.4	0.85
Mean		54.4	44.5	0.97
St Err		2.7	1.3	

Lowes Plot3 1991

Date	Wt 5	Tot. Samp	% Litt	R:S
23-04	0.11	40.5	47.4	1.27
	0.09	59.0	51.8	1.22
	0.09	59.2	39.2	1.13
	0.13	34.9	53.7	1.04
Mean	0.10	48.4	48.0	1.16
St Err	0.01	5.5	2.8	

Date	Wt 5	Tot. Samp	% Litt	R:S
25-06	0.06	7.7	60.4	0.77
	0.06	7.0	47.7	1.50
	0.09	18.1	58.4	1.22
	0.06	5.9	35.1	0.40
Mean	0.07	9.7	50.4	0.97
St Err	0.01	2.5	5.0	0.21

Date	Wt 5	Tot. Samp	% Litt	R:S
30-07	0.09	25.0	55.1	0.72
	0.07	14.9	89.2	1.04
	0.12	13.0	84.0	0.95
	0.06	5.9	46.0	1.75
Mean	0.08	14.7	68.6	1.12
St Err	0.01	3.4	9.2	

Date	Wt 5	Tot. Samp	% Litt	R:S
03-09	0.04	18.7	47.0	1.22
	0.08	22.1	65.2	1.00
	0.09	14.2	52.8	1.24
	0.07	20.6	57.4	0.97
Mean	0.07	18.9	55.6	1.11
St Err	0.01	1.5	3.3	

Table D.2 Species Composition in Study Sites 1991

Dee Plot 1
Date 9/4/91

Species	% Abundance (dry weight)			Mean	St Error
Elatine hexandra	0.0	0.3	0.0	0.0	0.1	0.1
Isoetes lacustris	1.4	36.1	5.0	1.9	11.1	8.4
Littorella uniflora	94.0	28.8	83.0	97.5	75.8	16.0
Lobelia dortmanna	4.3	34.8	12.0	0.6	12.9	7.7
Subularia aquatica	0.0	0.3	0.0	0.0	0.1	0.1

Date 4/6/91

Species	% Abundance (dry weight)			Mean	St Error
Elatine hexandra	1.0	7.2	0.0	0.3	2.1	1.7
Isoetes lacustris	5.8	6.3	13.4	0.0	6.4	2.7
Littorella uniflora	55.3	74.0	86.6	96.9	78.2	9.0
Lobelia dortmanna	28.2	4.3	0.0	2.2	8.7	6.6
Sphagnum sp.	2.9	0.0	0.0	0.0	0.7	0.7
Subularia aquatica	1.9	8.2	0.0	0.6	2.7	1.9

Date 9/7/91

Species	% Abundance (dry weight)			Mean	St Error
Isoetes lacustris	9.3	0.6	5.0	4.3	4.8	1.8
Littorella uniflora	90.7	98.5	83.0	95.7	92.0	3.4
Lobelia dortmanna	0.0	0.9	12.0	0.0	3.2	2.9

Date 13/8/91

Species	% Abundance (dry weight)			Mean	St Error
Elatine hexandra	0.0	0.0	0.0	0.5	0.1	0.1
Isoetes lacustris	54.9	29.1	38.8	38.5	40.3	5.4
Littorella uniflora	45.1	66.1	60.8	60.4	58.1	4.5
Lobelia dortmanna	0.0	4.3	0.0	0.0	1.1	1.1
Subularia aquatica	0.0	0.4	0.4	0.5	0.3	0.1

Dee Plot 2
Date 9/4/91

Species % Abundance (dry weight) Mean St Error
no samples avaliable

Date 4/6/91

Species	% Abundance (dry weight)			Mean	St Error
Isoetes lacustris	2.3	0.0	29.5	10.6	9.5
Littorella uniflora	94.5	100.0	70.5	88.3	9.1
Lobelia dortmanna	3.3	0.0	0.0	1.1	1.1

Date 9/7/91

Species	% Abundance (dry weight)			Mean	St Error
Isoetes lacustris	35.7	30.8	17.0	31.4	28.7	4.1
Littorella uniflora	42.9	50.8	83.0	68.6	61.3	9.0
Lobelia dortmanna	21.0	13.3	0.0	0.0	8.6	5.2
Subularia aquatica	0.4	5.0	0.0	0.0	1.4	1.2

Date 13/8/91

Species	% Abundance (dry weight)			Mean	St Error
Elatine hexandra	0.0	0.0	0.0	0.8	0.2	0.2
Isoetes lacustris	9.6	4.2	0.0	5.4	4.8	2.0
Littorella uniflora	88.5	95.8	100.0	93.6	94.5	2.4
Lobelia dortmanna	1.6	0.0	0.0	0.0	0.4	0.4
Subularia aquatica	0.3	0.0	0.0	2.6	0.7	0.6

Table D.2 (cont.) Species Composition in Study Sites 1991

Dee Plot 3
Date 9/4/91

Species	% Abundance (dry weight)			Mean	St Error
Isoetes lacustris	67.9	89.4	52.2	80.9	72.6	8.1
Littorella uniflora	26.5	7.8	36.3	11.0	20.4	6.7
Lobelia dortmanna	5.6	7.3	11.5	8.1	8.1	1.2
Subularia aquatica	0.0	0.0	8.8	0.0	2.2	2.2

Date 4/6/91

Species	% Abundance (dry weight)			Mean	St Error
Isoetes lacustris	31.3	19.3	73.6	35.7	40.0	11.7
Littorella uniflora	56.5	48.0	12.4	27.8	36.2	10.0
Lobelia dortmanna	11.6	32.7	14.0	36.5	23.7	6.4
Myriophyllum alterniflorum	0.7	0.0	0.0	0.0	0.2	0.2

Date 9/7/91

Species	% Abundance (dry weight)			Mean	St Error
Isoetes lacustris	28.8	0.0	58.0	72.7	39.9	16.1
Littorella uniflora	61.9	63.2	17.3	21.9	41.1	12.4
Lobelia dortmanna	9.3	6.6	22.2	5.5	10.9	3.9
Subularia aquatica	0.0	0.0	2.5	0.0	0.6	0.6

Date 13/8/91

Species	% Abundance (dry weight)			Mean	St Error
Isoetes lacustris	34.2	33.3	19.5	36.5	30.9	3.9
Littorella uniflora	39.5	50.0	64.1	58.1	52.9	5.3
Lobelia dortmanna	25.0	15.0	16.4	4.1	15.1	4.3
Subularia aquatica	1.3	1.7	0.0	1.4	1.1	0.4

Lomond Plot 1
Date 17/4/91

Species	% Abundance (dry weight)			Mean	St Error
Elodea sp	0.0	0.0	0.0	4.5	1.1	1.1
Juncus bulbosus	0.0	0.0	0.0	18.1	4.5	4.5
Littorella uniflora	100.0	100.0	100.0	77.5	94.4	5.6

Date 10/6/91

Species	% Abundance (dry weight)			Mean	St Error
Littorella uniflora	100.0	99.1	100.0	100.0	99.8	0.2
Myriophyllum alterniflorum	0.0	0.9	0.0	0.0	0.2	0.2

Date 18/7/91

Species	% Abundance (dry weight)			Mean	St Error
Juncus bulbosus	0.0	0.0	5.1	0.0	1.3	1.3
Littorella uniflora	100.0	100.0	94.9	100.0	98.7	1.3

Date 20/8/91

Species	% Abundance (dry weight)			Mean	St Error
Littorella uniflora	100.0	97.4	98.2	99.1	98.7	0.6
Myriophyllum alterniflorum	0.0	2.6	0.7	0.2	0.9	0.6
Potamogeton crispus	0.0	0.0	0.1	0.7	0.2	0.2

Table D.2 (cont.) Species Composition in Study Sites 1991

Lomond Plot 2
Date 17/4/91

Species	% Abundance (dry weight)				Mean	St Error
Littorella uniflora	100.0	100.0	100.0	100.0	100.0	0.0

Date 10/6/91

Species	% Abundance (dry weight)				Mean	St Error
Elodea canadensis	0.0	0.0	0.3	0.0	0.1	0.1
Isoetes lacustris	4.4	4.4	3.7	2.4	3.7	0.5
Littorella uniflora	92.9	79.2	94.1	97.6	91.0	4.0
Lobelia dortmanna	0.7	16.3	2.0	0.0	4.8	3.9

Date 18/7/91

Species	% Abundance (dry weight)				Mean	St Error
Elodea canadensis	0.0	0.2	0.0	2.5	0.7	0.6
Littorella uniflora	100.0	99.8	100.0	97.5	99.3	0.6

Date 20/8/91

Species	% Abundance (dry weight)				Mean	St Error
Elodea canadensis	0.0	0.0	3.2	0.0	0.8	0.8
Isoetes lacustris	0.2	0.1	13.9	4.7	4.7	3.2
Littorella uniflora	99.8	97.4	82.9	95.3	93.9	3.8
Lobelia dortmanna	0.0	2.5	0.0	0.0	0.6	0.6

Lomond Plot 3
Date 17/4/91

Species	% Abundance (dry weight)				Mean	St Error
Littorella uniflora	100.0	100.0	100.0	97.1	99.3	0.7
Lobelia dortmanna	0.0	0.0	0.0	2.9	0.7	0.7

Date 10/6/91

Species	% Abundance (dry weight)				Mean	St Error
Elodea canadensis	0.0	0.6	0.0	0.0	0.2	0.2
Isoetes lacustris	0.0	4.7	0.0	0.0	1.2	1.2
Littorella uniflora	100.0	94.7	100.0	100.0	98.7	1.3

Date 18/7/91

Species	% Abundance (dry weight)				Mean	St Error
Isoetes lacustris	0.0	0.0	5.8	0.0	1.5	1.5
Littorella uniflora	100.0	96.8	94.2	100.0	97.8	1.4
Lobelia dortmanna	0.0	3.2	0.0	0.0	0.8	0.8

Date 20/8/91

Species	% Abundance (dry weight)				Mean	St Error
Elodea canadensis	0.3	0.5	1.4	0.0	0.6	0.3
Isoetes lacustris	7.2	2.0	0.5	3.0	3.2	1.4
Littorella uniflora	92.1	90.4	95.8	85.7	91.0	2.1
Lobelia dortmanna	0.0	7.1	2.3	10.5	5.0	2.4
Potamogeton sp.	0.3	0.0	0.0	0.6	0.2	0.1

Table D.2 (cont.) Species Composition in Study Sites 1991

Menteith Plot 1
Date 1/5/91

Species	% Abundance (dry weight)			Mean	St Error
Nitella translucens	0.2	1.9	4.0	0.2	1.6	0.9
Isoetes lacustris	83.8	53.8	89.9	62.4	72.5	8.6
Littorella uniflora	12.1	42.3	6.1	37.3	24.5	9.0
Myriophyllum alterniflorum	4.0	1.9	0.0	0.0	1.5	1.0

Date 18/6/91

Species	% Abundance (dry weight)			Mean	St Error
Nitella translucens	0.0	0.2	1.5	0.0	0.4	0.4
Isoetes lacustris	61.1	84.0	67.6	94.6	76.8	7.6
Littorella uniflora	38.9	15.8	31.0	5.4	22.8	7.5

Date 23/7/91

Species	% Abundance (dry weight)			Mean	St Error
Nitella translucens	0.0	0.0	0.0	0.0	0.0	0.0
Isoetes lacustris	85.2	94.6	13.2	37.1	57.5	19.4
Littorella uniflora	14.8	5.4	85.6	57.6	40.9	18.7
Myriophyllum alterniflorum	0.0	0.0	1.2	5.3	1.6	1.3

Date 27/8/91

Species	% Abundance (dry weight)			Mean	St Error
Isoetes lacustris	47.7	77.0	1.7	33.6	40.0	15.6
Littorella uniflora	45.4	21.0	93.8	64.3	56.1	15.4
Myriophyllum alterniflorum	7.0	1.9	4.5	2.1	3.9	1.2

Menteith Plot 2
Date 1/5/91

Species	% Abundance (dry weight)			Mean	St Error
Nitella translucens	0.0	0.0	0.0	2.2	0.6	0.6
Isoetes lacustris	27.7	39.8	19.7	24.9	28.0	4.3
Littorella uniflora	72.0	54.1	78.0	71.3	68.9	5.1
Lobelia dortmanna	0.3	6.1	2.3	1.6	2.6	1.2

Date 18/6/91

Species	% Abundance (dry weight)			Mean	St Error
Nitella translucens	0.6	0.6	0.0	13.1	3.6	3.2
Isoetes lacustris	5.1	0.6	0.9	21.1	6.9	4.8
Littorella uniflora	93.7	98.8	92.8	63.6	87.2	8.0
Lobelia dortmanna	0.0	0.0	0.0	0.0	0.0	0.0
Myriophyllum alterniflorum	0.0	0.0	5.9	0.0	1.5	1.5
Potamogeton gramineus	0.6	0.0	0.5	0.0	0.3	0.2

Date 23/7/91

Species	% Abundance (dry weight)			Mean	St Error
Nitella translucens	0.0	1.3	0.0	0.9	0.6	0.3
Elodea canadensis	0.0	1.9	0.4	0.0	0.6	0.5
Isoetes lacustris	11.8	15.3	8.9	6.2	10.6	2.0
Littorella uniflora	88.2	81.8	90.7	93.8	88.6	2.5
Myriophyllum alterniflorum	0.0	0.0	0.0	0.0	0.0	0.0

Date 27/8/91

Species	% Abundance (dry weight)			Mean	St Error
Elodea canadensis	0.0	0.0	0.0	0.6	0.2	0.2
Isoetes lacustris	28.7	19.0	16.4	17.3	20.4	2.8
Littorella uniflora	70.6	71.5	83.6	82.1	77.0	3.4
Myriophyllum alterniflorum	0.0	9.5	0.0	0.0	2.4	2.4
Potamogeton perfoliatus	0.7	0.0	0.0	0.0	0.2	0.2

Table D.2 (cont.) Species Composition in Study Sites 1991

Menteith Plot 3

Date 1/5/91

Species	% Abundance (dry weight)			Mean	St Error
Nitella translucens	0.2	0.2	3.0	0.2	0.9	0.7
Isoetes lacustris	86.3	90.5	94.7	90.4	90.5	1.7
Littorella uniflora	13.5	9.3	2.3	9.1	8.6	2.3
Lobelia dortmanna	0.0	0.0	0.0	0.4	0.1	0.1

Date 18/6/91

Species	% Abundance (dry weight)			Mean	St Error
Nitella translucens	2.3	0.0	3.6	76.3	20.6	18.6
Isoetes lacustris	61.7	74.4	66.4	23.7	56.6	11.3
Littorella uniflora	36.0	25.6	30.0	0.0	22.9	7.9

Date 23/7/91

Species	% Abundance (dry weight)			Mean	St Error
Nitella translucens	0.0	4.5	1.0	1.8	1.4
Isoetes lacustris	72.5	72.7	75.1	73.4	0.8
Littorella uniflora	27.5	22.8	23.9	24.7	1.4

Date 13/8/91

Species	% Abundance (dry weight)			Mean	St Error
Isoetes lacustris	85.3	71.9	89.8	85.1	83.0	3.9
Littorella uniflora	14.7	28.1	10.2	14.9	17.0	3.9

Lowes Plot 1

Date 23/4/91

Species	% Abundance (dry weight)				Mean	St Error
Chara globularis	5.6	11.5	21.4	23.6	15.5	4.2
Elodea canadensis	0.0	0.6	0.0	0.0	0.2	0.2
Isoetes lacustris	9.3	19.4	13.8	9.5	13.0	2.4
Juncus bulbosus	19.9	27.9	26.5	26.8	25.3	1.8
Littorella uniflora	65.2	40.6	38.4	40.1	46.1	6.4

Date 25/6/91

Species	% Abundance (dry weight)				Mean	St Error
Chara globularis	44.5	5.6	2.2	5.2	14.4	10.1
Isoetes lacustris	16.1	7.6	16.8	12.9	13.4	2.1
Juncus bulbosus	0.0	18.5	4.9	2.6	6.5	4.1
Littorella uniflora	39.4	68.2	76.2	79.4	65.8	9.1

Date 30/7/91

Species	% Abundance (dry weight)					Mean	St Error
Chara globularis	0.4	0.0	1.1	0.9	0.6	0.2
Elatine hexandra	0.0	0.4	0.0	0.9	0.3	0.2
Elodea canadensis	0.4	0.0	0.0	0.0	0.1	0.1
Isoetes lacustris	34.4	26.4	0.0	16.5	19.3	7.4
Juncus bulbosus	22.4	22.0	19.8	2.8	16.8	4.7
Littorella uniflora	42.3	44.0	79.1	72.5	59.5	9.5
Lobelia dortmanna	0.0	0.0	0.0	6.4	1.6	1.6
Myriophyllum alterniflorum	0.0	7.2	0.0	0.0	1.8	1.8
Potamogeton gramineus	0.8	0.0	0.0	0.0	0.2	0.2

Date 13/8/91

Species	% Abundance (dry weight)				Mean	St Error
Chara globularis	1.4	8.4	2.8	8.5	5.3	1.9
Elatine hexandra	0.0	0.0	0.7	0.0	0.2	0.2
Isoetes lacustris	14.4	42.1	22.2	15.3	23.5	6.4
Juncus bulbosus	15.5	15.9	7.6	5.1	11.0	2.7
Littorella uniflora	68.6	33.6	66.7	71.2	60.0	8.9

Table D.2 (cont.) Species Composition in Study Sites 1991

Lowes Plot 2

Date 23/4/91

Species	% Abundance (dry weight)			Mean	St Error
Chara globularis	5.9	12.1	5.2	7.1	1.7
Isoetes lacustris	24.1	15.0	18.1	18.6	1.9
Juncus bulbosus	12.7	3.8	16.5	12.0	2.8
Littorella uniflora	57.3	69.0	60.2	62.2	2.5
Subularia aquatica	0.0	0.0	0.0	0.1	0.1

Date 25/6/91

Species	% Abundance (dry weight)			Mean	St Error
Chara globularis	3.3	1.4	10.5	3.8	2.3
Isoetes lacustris	23.7	27.1	25.5	21.6	3.9
Juncus bulbosus	38.2	1.4	13.7	27.6	12.4
Littorella uniflora	33.6	65.7	50.3	45.6	7.8
Potamogeton gramineus	0.0	4.3	0.0	1.1	1.1
Subularia aquatica	1.3	0.0	0.0	0.3	0.3

Date 30/7/91

Species	% Abundance (dry weight)			Mean	St Error
Chara globularis	0.0	0.0	0.6	1.9	1.7
Isoetes lacustris	8.8	2.5	9.9	7.8	1.8
Juncus bulbosus	0.0	1.2	0.6	1.0	0.5
Littorella uniflora	91.3	96.3	88.3	89.3	3.2
Myriophyllum alterniflorum	0.0	0.0	0.6	0.2	0.2

Date 13/8/91

Species	% Abundance (dry weight)			Mean	St Error
Chara globularis	0.0	1.4	0.0	0.4	0.4
Isoetes lacustris	2.0	9.4	22.7	20.7	10.3
Juncus bulbosus	17.6	16.7	5.5	16.9	4.5
Littorella uniflora	80.4	60.1	53.6	54.4	11.8
Myriophyllum alterniflorum	0.0	12.3	18.2	7.6	4.6

Lowes Plot 3

Date 23/4/91

Species	% Abundance (dry weight)			Mean	St Error
Chara globularis	20.6	0.4	2.7	8.3	4.5
Isoetes lacustris	26.5	0.6	8.4	13.9	5.8
Juncus bulbosus	5.5	97.0	49.6	42.1	20.6
Littorella uniflora	47.4	2.0	39.1	35.6	11.6
Subularia aquatica	0.0	0.0	0.3	0.2	0.1

Date 25/6/91

Species	% Abundance (dry weight)			Mean	St Error
Chara globularis	32.0	2.3	0.0	8.6	7.8
Isoetes lacustris	13.7	2.3	10.8	14.8	6.3
Juncus bulbosus	41.1	47.7	31.5	37.5	4.2
Littorella uniflora	13.2	47.7	57.7	38.4	9.6
Myriophyllum alterniflorum	0.0	0.0	0.0	0.7	0.7

Date 30/7/91

Species	% Abundance (dry weight)			Mean	St Error
Chara globularis	0.6	0.0	0.0	0.8	0.6
Elodea canadensis	1.9	0.0	0.0	0.5	0.5
Isoetes lacustris	12.8	2.2	7.4	8.3	2.3
Juncus bulbosus	29.5	8.6	8.6	21.8	7.9
Littorella uniflora	55.1	89.2	84.0	68.6	10.6

Date 13/8/91

Species	% Abundance (dry weight)			Mean	St Error
Chara globularis	0.0	4.8	6.7	4.2	1.5
Elatine hexandra	0.0	0.7	0.0	0.4	0.2
Isoetes lacustris	12.0	9.0	27.0	16.5	4.0
Juncus bulbosus	41.0	23.4	13.5	22.9	6.5
Littorella uniflora	47.0	62.1	52.8	53.7	3.1

Figure D2b.i Macrophyte % Abundance Dry Weight
1991 Loch Dee Plot 1

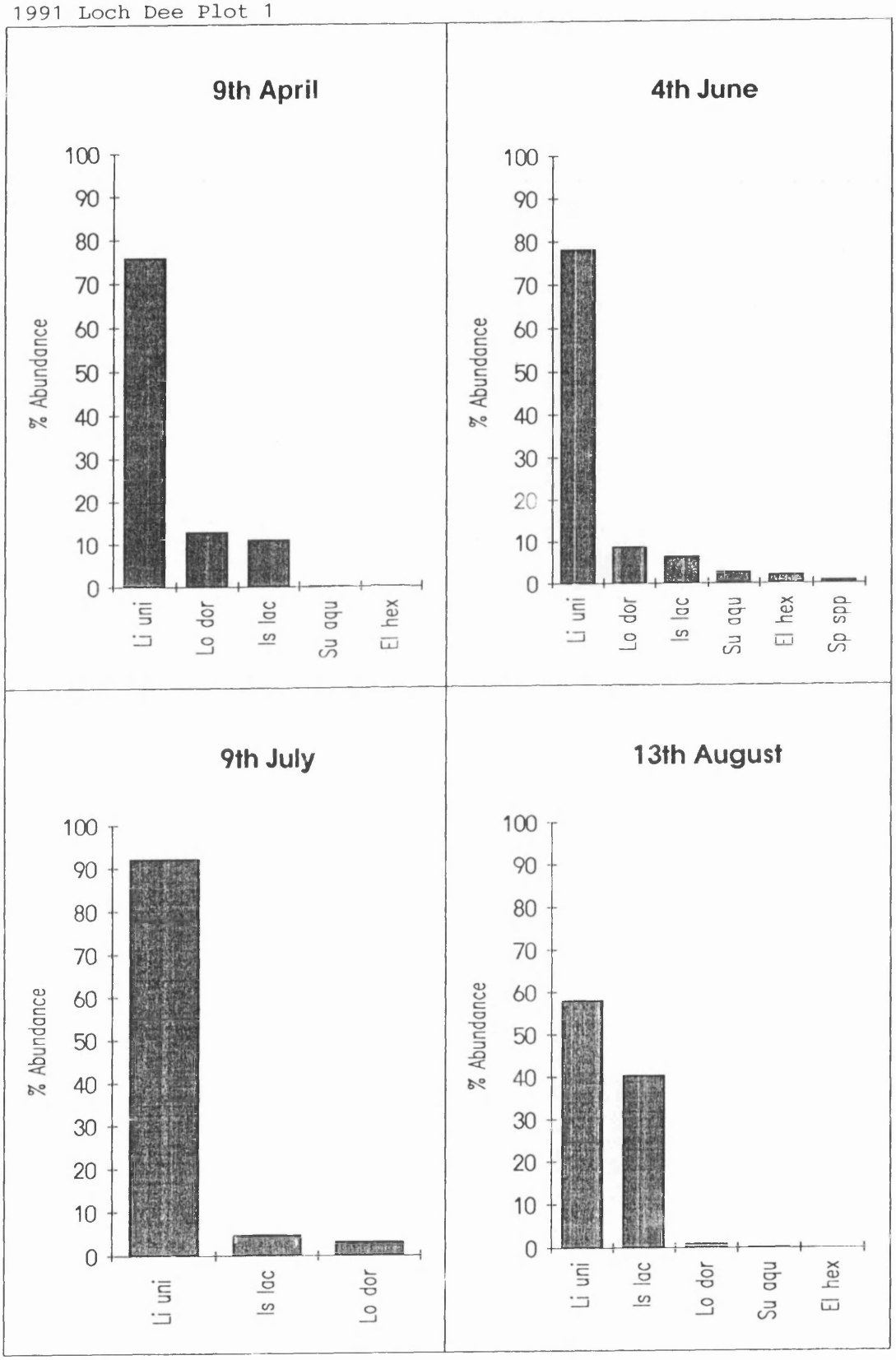


Figure D2b.ii Macrophyte % Abundance Dry Weight
1991 Loch Dee Plot 2

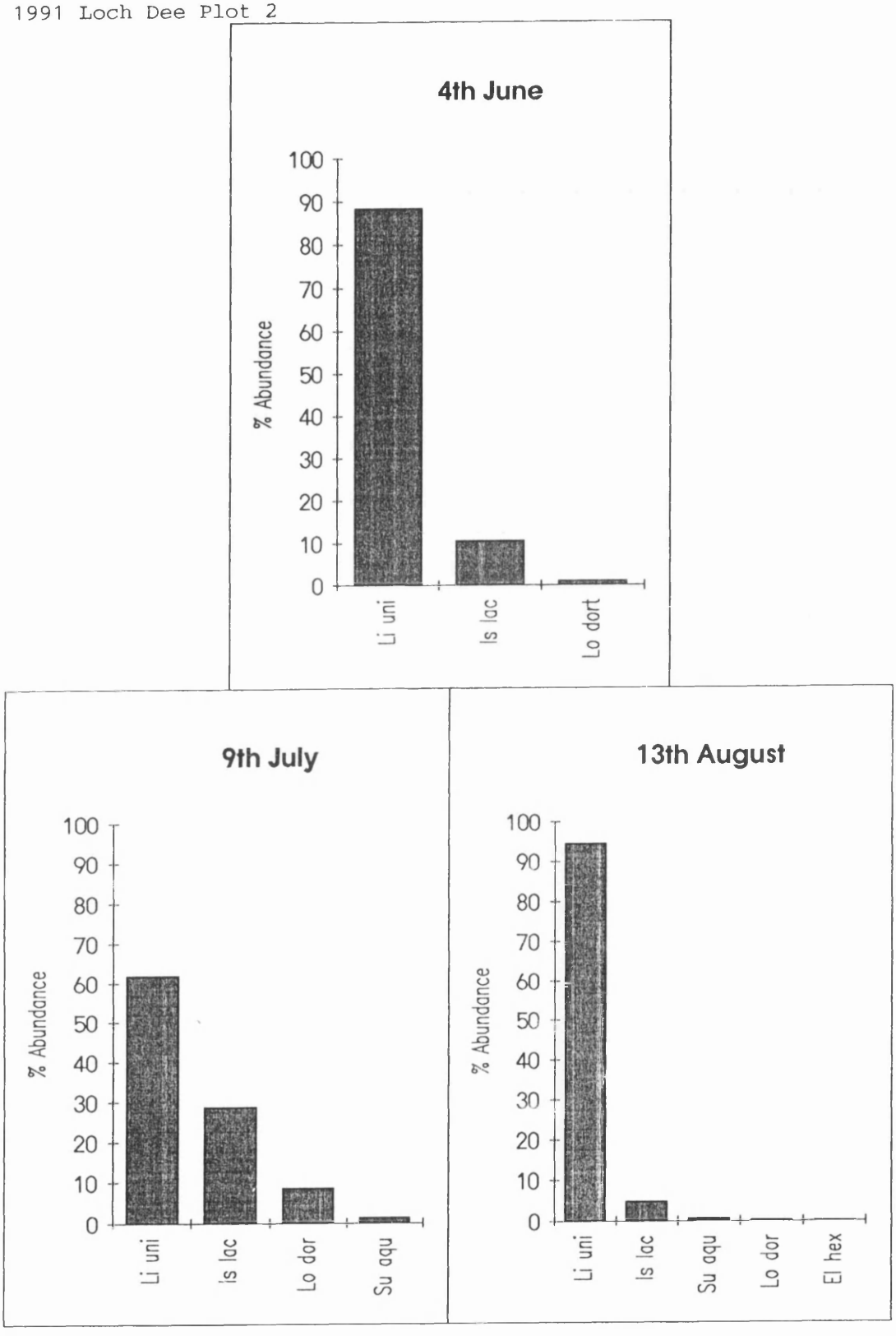


Figure D2b.iii Macrophyte % Abundance Dry Weight
1991 Loch Dee Plot 3

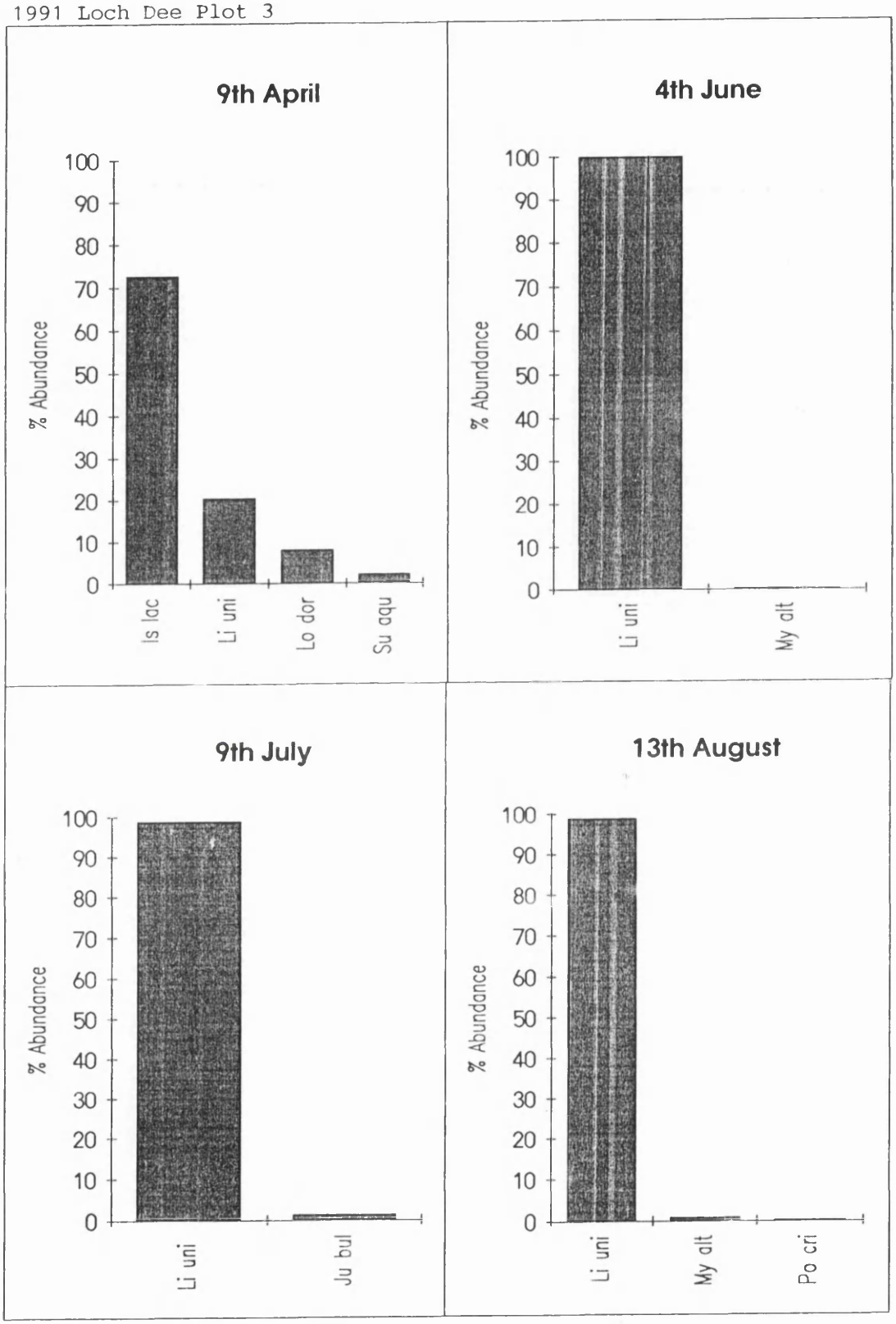


Figure D2b.iv Macrophyte % Abundance Dry Weight
1991 Loch Lomond Plot 1

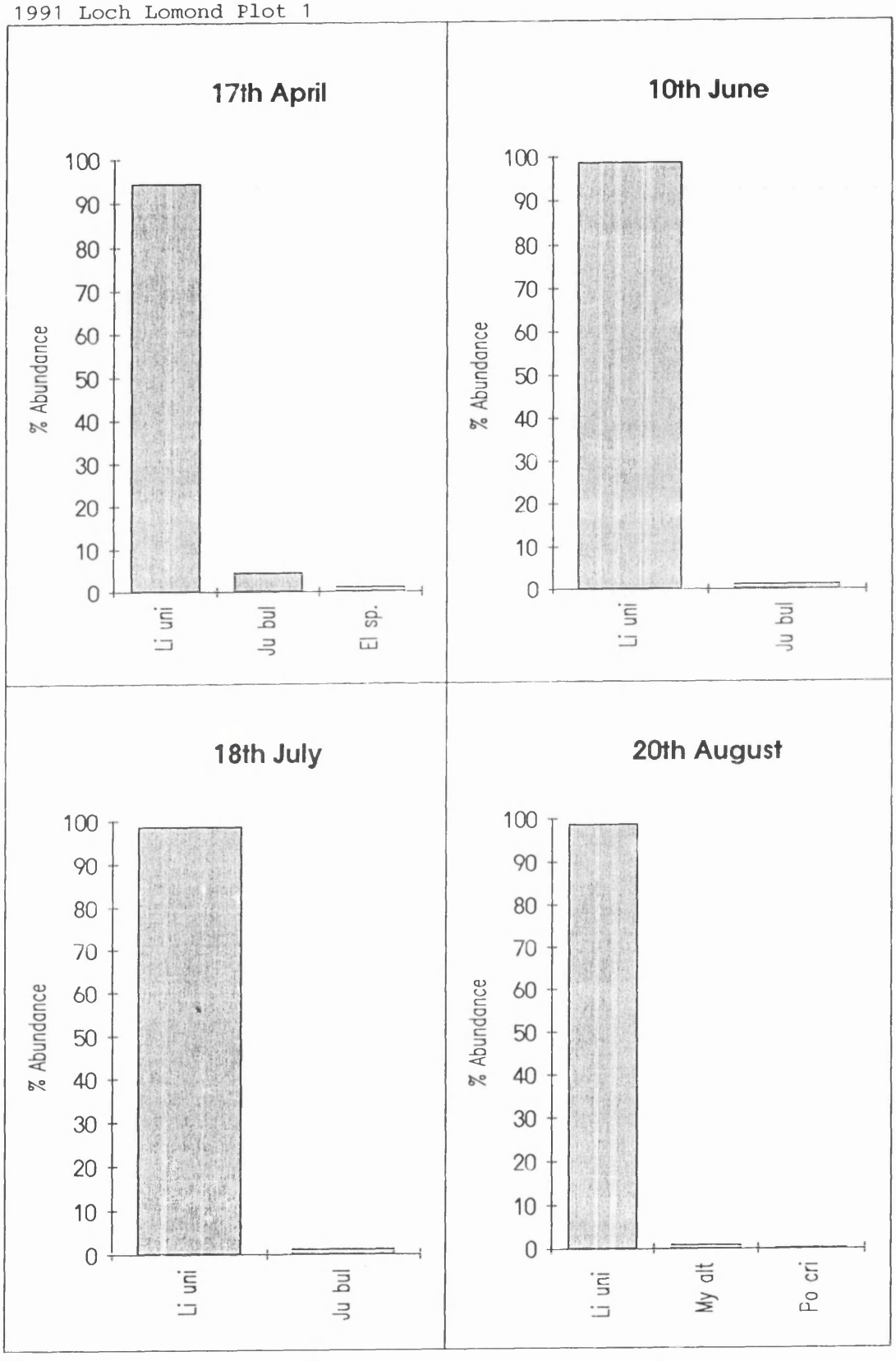


Figure D2b.v Macrophyte % Abundance Dry Weight
1991 Loch Lomond Plot 2

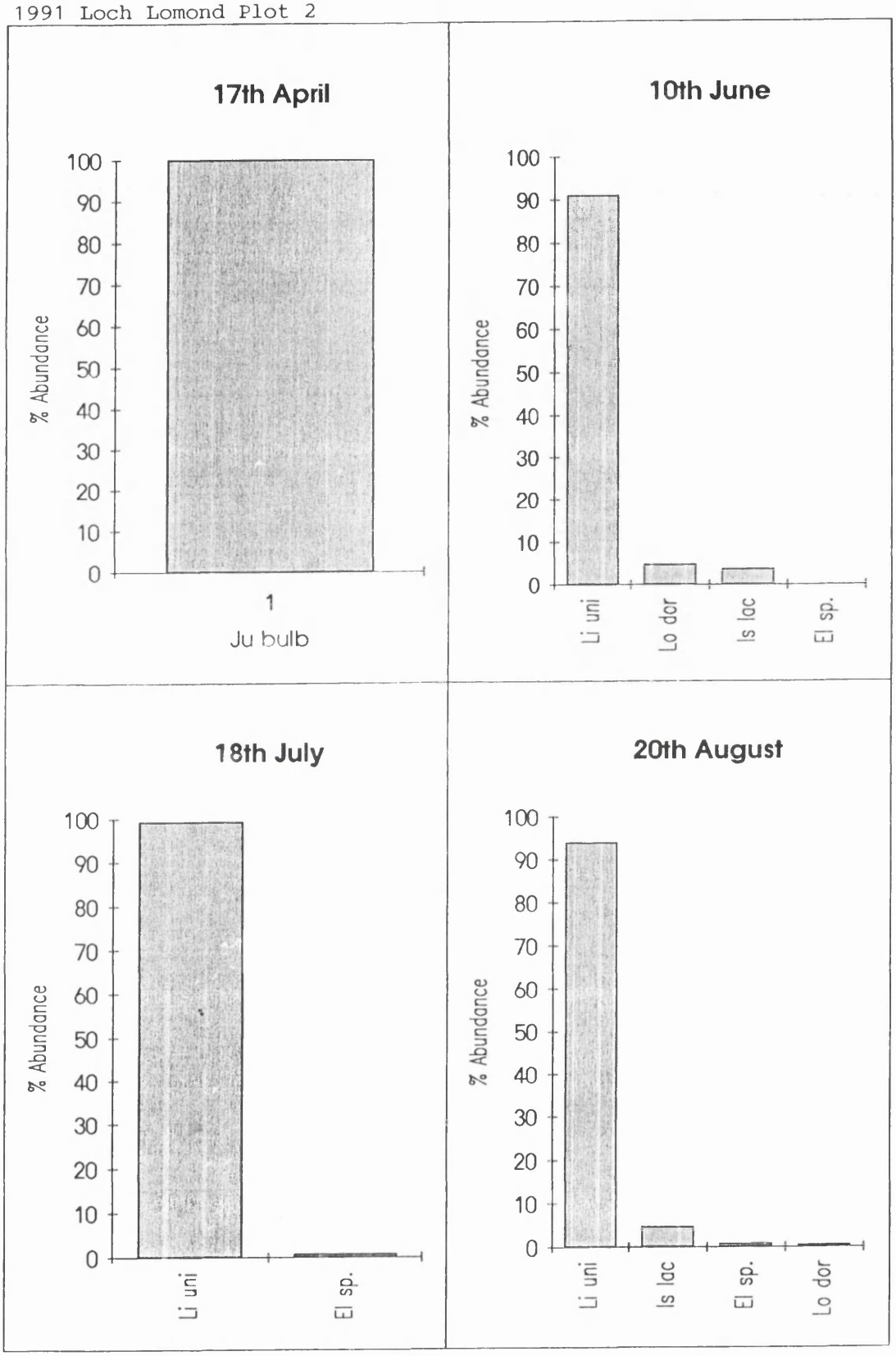


Figure D2b.vi Macrophyte % Abundance Dry Weight
1991 Loch Lomond Plot 3

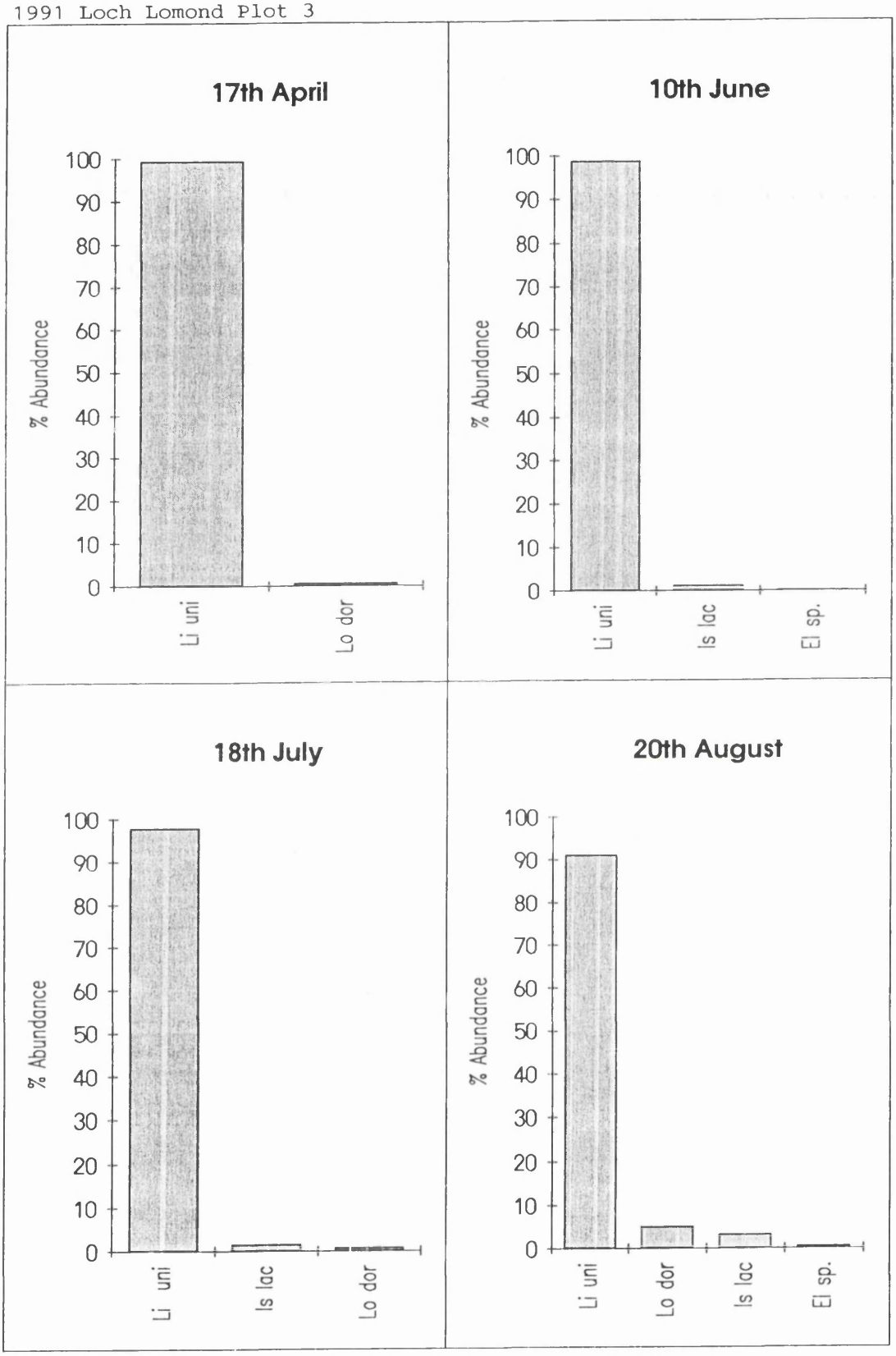


Figure D2b.vii Macrophyte % Abundance Dry Weight
1991 Lake of Menteith Plot 1

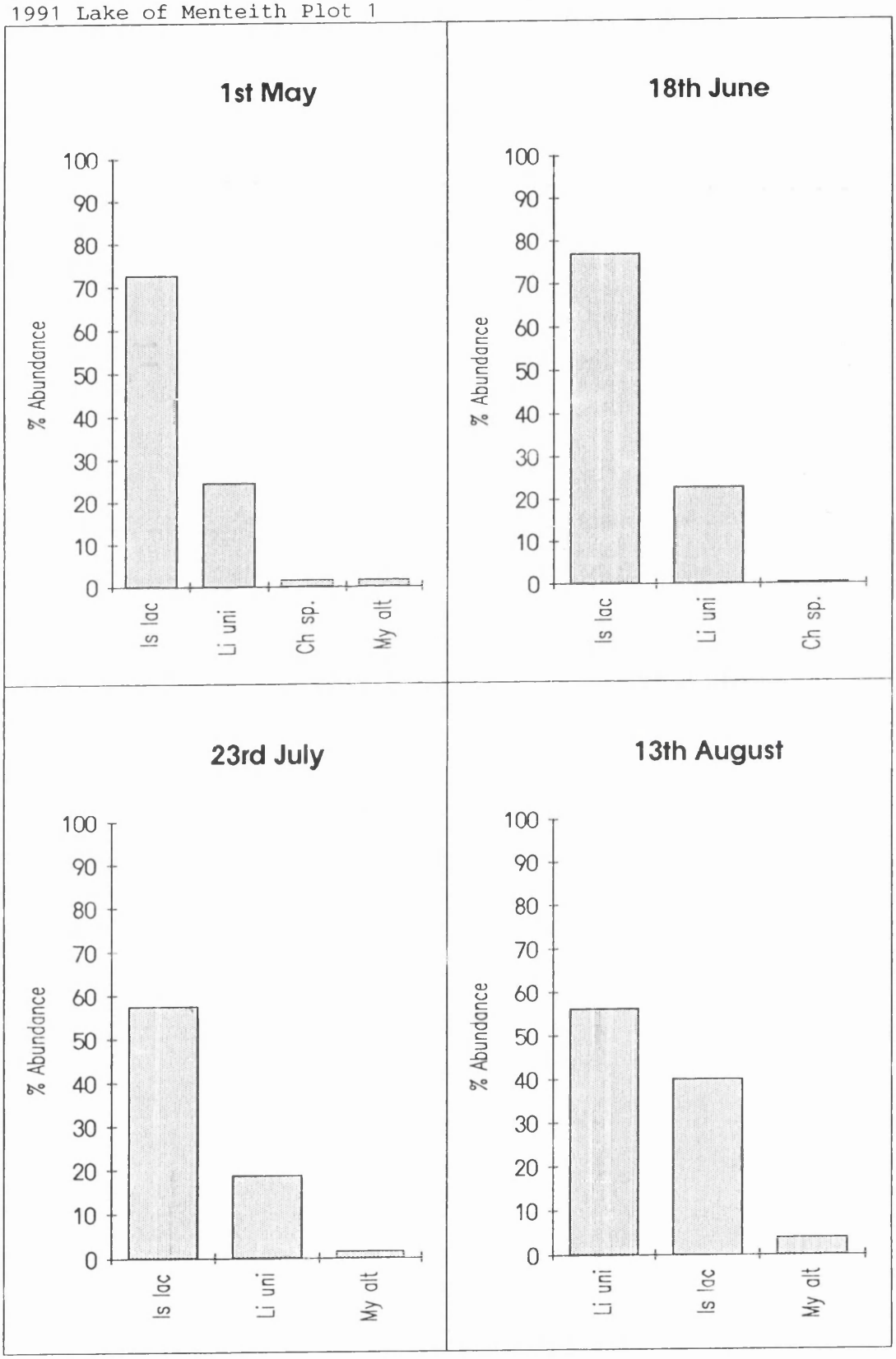


Figure D2b.viii Macrophyte % Abundance dry Weight
1991 Lake of Menteith Plot 2

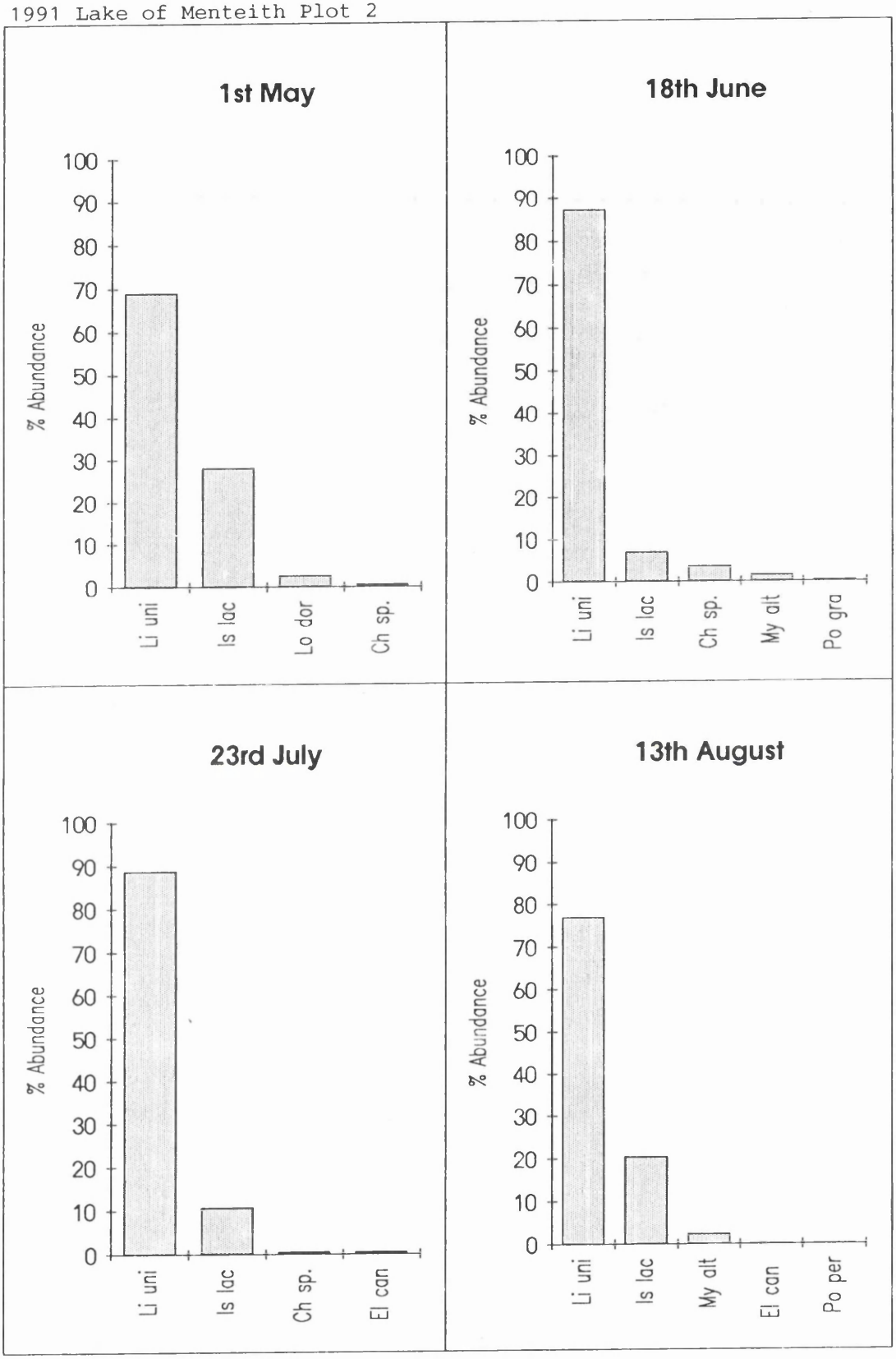


Figure D2b.ix Macrophyte % Abundance Dry Weight
1991 Lake of Menteith Plot 3

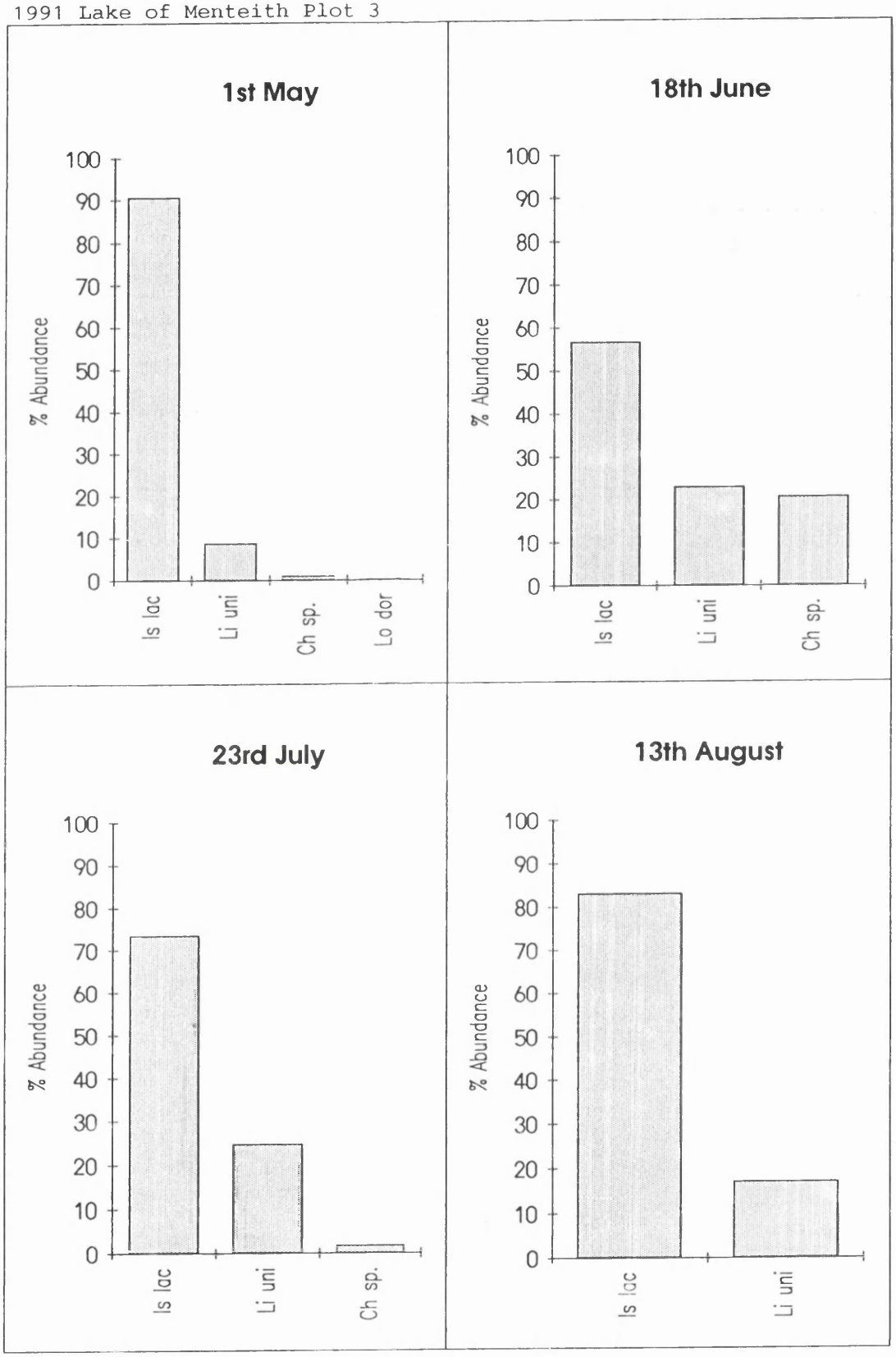


Figure D2b.x Macrophyte % Abundance Dry Weight
1991 Loch of Lowes Plot 1

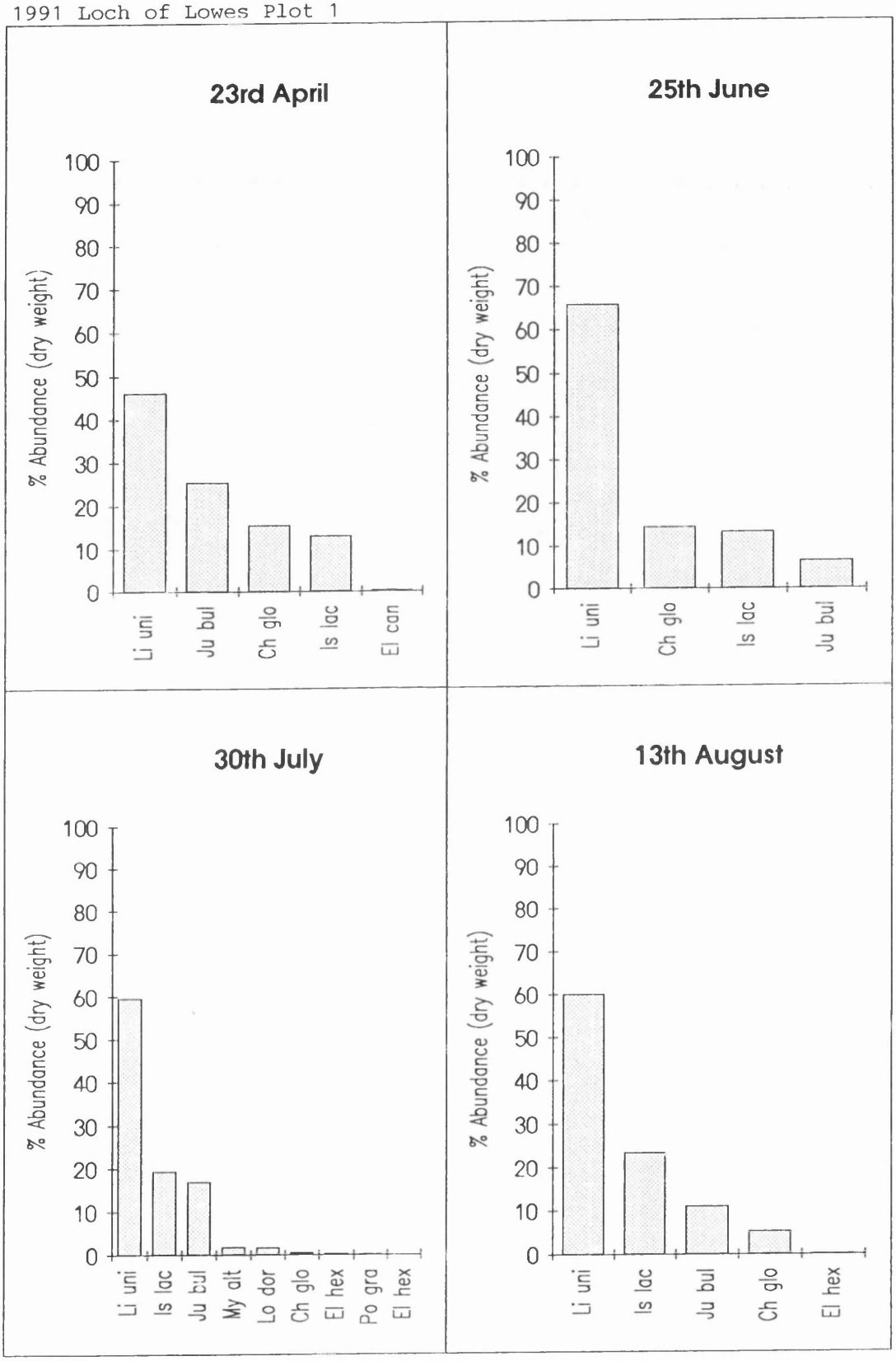
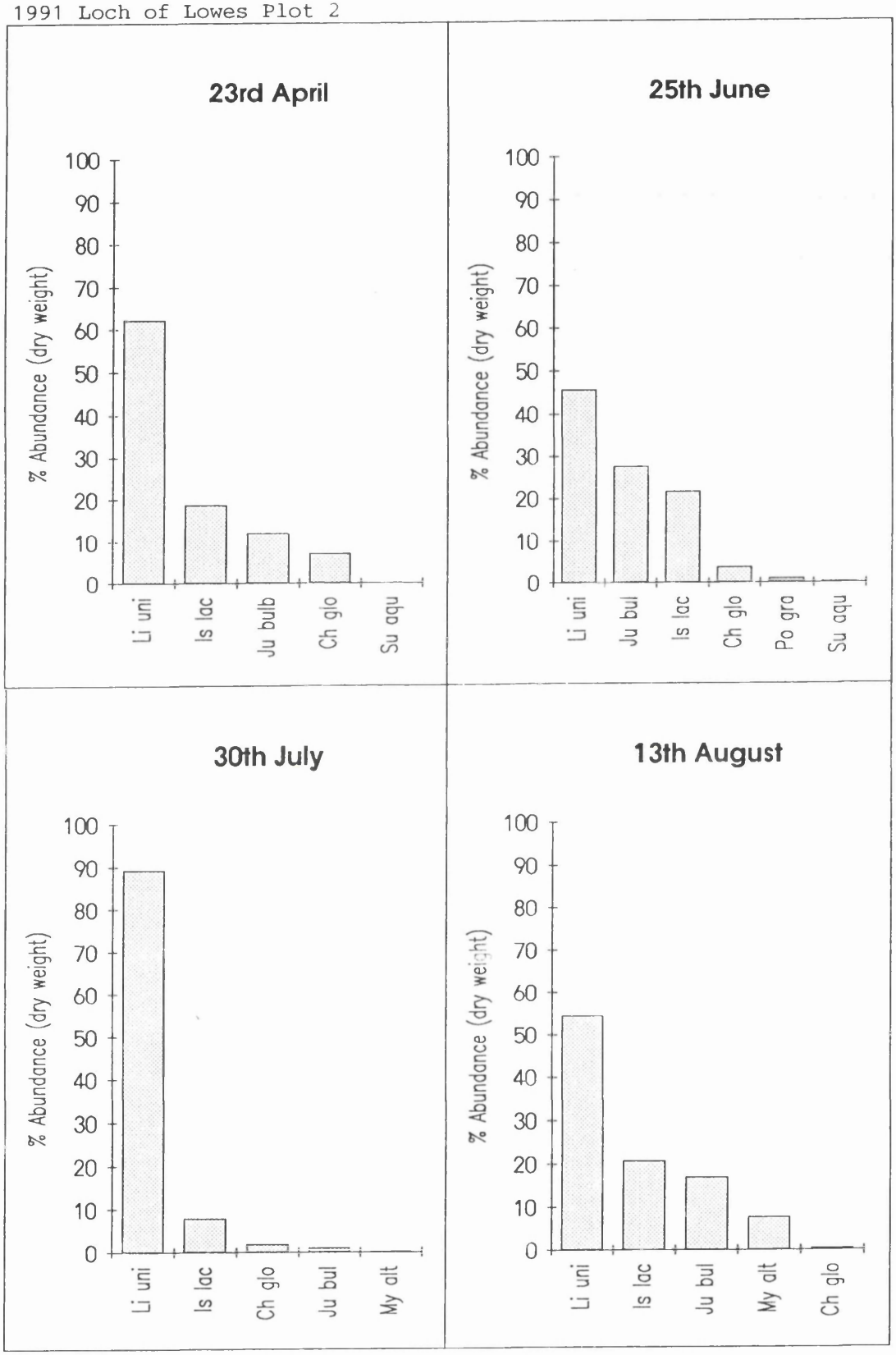
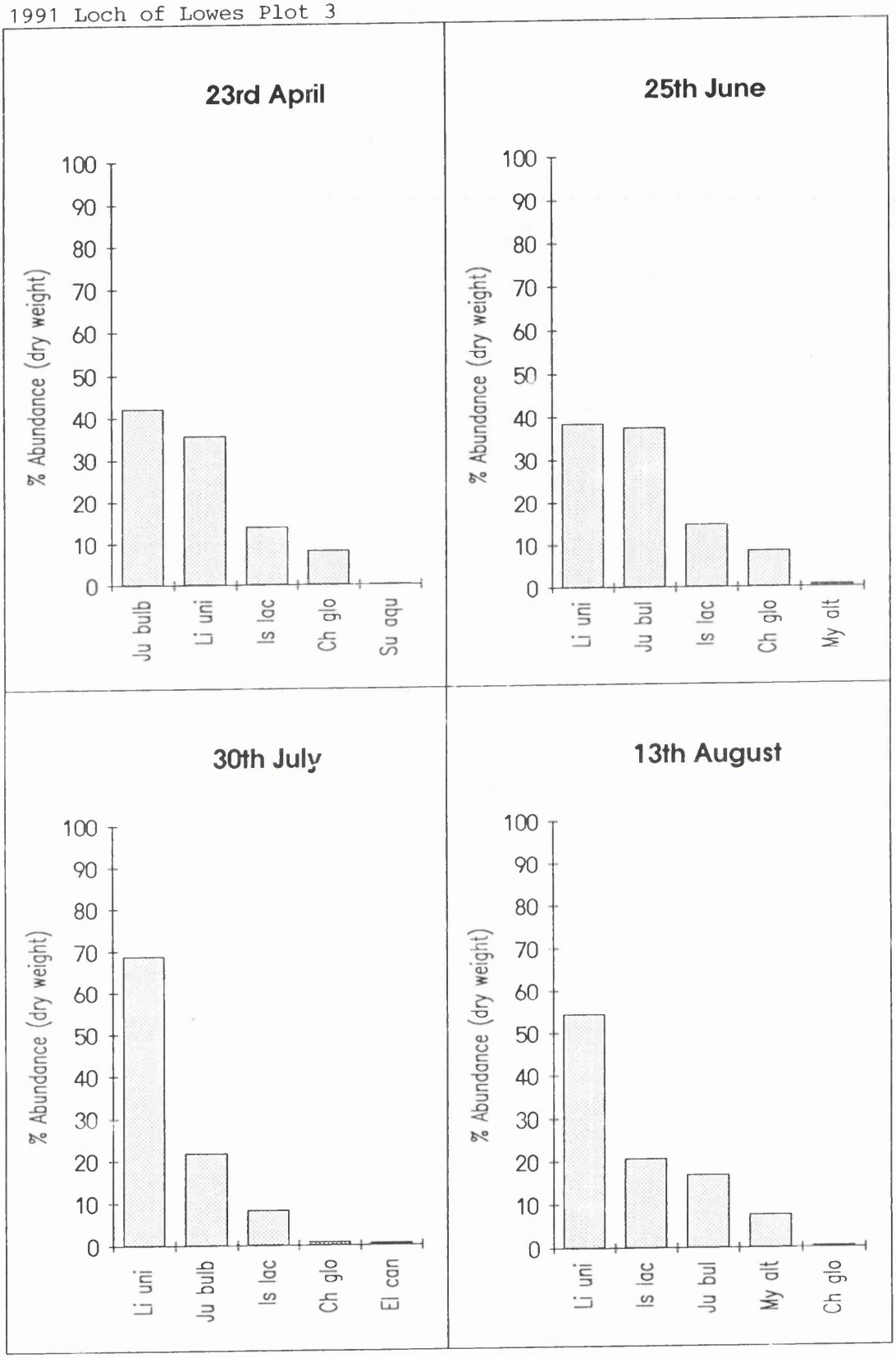


Figure D2b.xi Macrophyte % Abundance Dry Weight
1991 Loch of Lowes Plot 2



FigureD2b.xii Macrophyte % Abundance Dry Weight
1991 Loch of Lowes Plot 3



Appendix D3a
Littorella Attribute data used in Stepwise multiple regression

01	96	5.5	32	1.15	74	14.1	69	50
02	82	4.9	32	1.12	67	14.3	67	74
03	81	4.1	35	1.07	71	15.7	71	55
04	106	5.3	51	0.89	92	13.0	128	175
05	88	5.1	37	1.56	64	14.8	98	63
06	94	4.9	32	1.36	62	16.0	108	67
07	85	5.1	44	1.18	96	15.2	142	137
08	75	5.3	35	1.33	97	15.7	143	139
09	90	5.7	37	1.30	99	13.8	151	140
10	88	4.1	32	0.83	90	18.4	42	38
11	93	4.2	48	1.28	97	10.3	158	153
12	56	4.2	29	0.87	12	12.3	22	3
13	92	4.2	37	1.14	44	17.0	44	9
14	114	4.2	54	1.19	88	17.9	107	81
15	72	4.4	27	1.17	59	18.8	40	11
16	92	4.2	37	0.93	68	17.7	43	24
17	94	4.3	45	1.08	87	14.1	125	117
18	65	4.1	25	1.13	28	15.6	22	6
19	75	6.0	27	0.99	70	10.6	38	27
20	81	5.5	22	1.19	37	18.9	42	16
21	90	4.5	21	0.76	19	18.8	42	8
22	76	5.5	25	1.13	68	16.9	58	40
23	81	4.3	29	1.33	42	21.2	61	44
24	80	3.9	33	1.08	7	22.3	389	26
25	79	6.1	26	1.35	86	14.8	81	70
26	88	4.9	26	1.55	91	16.6	100	92
27	99	4.3	55	0.94	10	15.4	381	39
28	68	3.8	33	1.00	37	21.5	19	7
29	65	4.6	21	1.06	45	18.9	22	10
30	73	4.7	22	1.08	70	24.2	14	10
31	67	3.8	35	1.37	60	17.1	53	25
32	87	3.8	50	1.31	56	16.8	57	32
33	92	3.8	56	1.16	25	16.9	75	19
34	67	4.0	37	1.69	43	15.5	71	30
35	75	4.1	45	1.17	44	17.6	56	25
36	79	4.3	25	0.97	44	15.4	54	24

00

STOLONL	LEAFNO	LEAFL	ROOTSH	PERABUN	LFCHL	TOTBIO	LITTBIO		
01LOM21	02LOM22	03LOM23	04LOM31	05LOM32	06LOM33	07LOM41	08LOM42	09LOM43	10DEE21
11DEE22	12DEE23	13DEE31	14DEE32	15DEE33	16DEE41	17DEE42	18DEE43	19MEN21	20MEN22
21MEN23	22MEN31	23MEN32	24MEN33	25MEN41	26MEN42	27MEN43	28LOW21	29LOW22	30LOW23
31LOW31	32LOW32	33LOW33	34LOW41	35LOW42	36LOW42				

Appendix D3b
Data used in Stepwise Linear Mulitple Regression 1991

01	0.25	105	13.7	1132	0.23	1.00	76	3.2	3.9	46	43	33
02	0.21	107	20.6	1516	0.15	0.76	79	3.5	5.1	52	47	37
03	0.24	98	14.9	814	0.42	1.21	92	3.2	4.7	51	54	49
04	0.31	105	14.0	1065	0.11	0.69	58	5.3	4.0	47	38	22
05	*	*	*	*	*	*	*	*	*	*	*	*
06	0.06	102	22.4	1814	0.15	1.54	88	3.3	4.7	49	59	52
07	0.93	102	18.7	928	0.13	0.62	61	3.7	4.3	49	39	28
08	0.29	128	14.3	2277	0.15	0.70	94	4.5	4.7	55	70	67
09	0.24	68	13.4	645	0.05	0.92	20	2.9	4.0	32	40	8
10	0.10	64	19.6	567	0.05	0.90	31	3.7	4.9	23	26	8
11	0.42	74	16.7	539	0.06	0.96	41	2.1	4.6	17	16	7
12	0.36	66	10.6	830	0.05	1.03	53	3.7	5.4	18	14	7
13	0.38	70	11.5	1602	0.17	0.98	94	3.0	5.6	43	58	54
14	0.11	72	9.6	3352	0.16	1.04	99	2.8	5.8	39	112	111
15	0.17	70	10.8	4232	0.23	1.15	98	2.9	6.1	42	215	210
16	0.09	64	11.4	252	0.26	0.84	99	2.9	5.8	46	146	114
17	0.13	56	11.5	5378	0.17	0.93	100	3.2	6.0	38	186	186
18	0.06	71	9.6	2603	0.14	1.32	91	2.0	5.2	27	74	68
19	0.18	80	10.8	2068	0.19	1.17	99	2.7	5.8	36	90	89
20	0.23	63	10.0	568	0.23	1.20	95	2.1	5.4	36	118	112
21	0.20	63	11.5	4235	0.19	0.82	99	3.1	5.9	34	155	154
22	0.04	62	10.8	5898	0.15	1.05	99	2.8	6.2	35	140	138
23	0.20	70	10.8	3200	0.13	1.38	97	2.6	5.3	30	111	108
24	0.25	89	10.4	748	0.21	1.23	91	2.5	5.5	41	136	124
25	0.40	55	14.5	738	0.11	0.59	24	2.7	5.4	27	73	18
26	0.19	74	14.5	638	0.09	1.04	23	3.0	5.4	27	65	15
27	0.10	68	14.8	1698	0.08	0.92	41	2.8	5.4	28	92	38
28	0.19	68	15.5	638	0.09	1.08	56	3.0	5.6	24	52	29
29	0.08	57	13.7	3062	0.08	1.45	69	3.1	6.1	20	66	45
30	0.12	68	12.8	1559	0.09	1.25	87	3.2	5.8	26	33	28
31	0.10	58	13.0	1801	0.13	1.34	89	2.9	5.9	30	54	48
32	0.12	88	12.6	1559	0.12	0.96	67	4.0	5.1	41	60	41
33	0.04	59	11.7	688	0.05	0.86	8	3.2	3.7	29	87	7
34	0.31	73	10.9	1583	0.13	1.04	29	4.5	5.9	42	141	41
35	0.50	95	11.7	1075	0.14	1.29	25	3.6	5.1	45	109	27
36	0.31	97	12.5	1583	0.17	0.73	17	3.6	5.7	51	263	45
37	0.13	59	15.0	713	0.12	0.84	46	2.7	5.1	30	39	18
38	0.25	66	20.6	1281	0.08	1.25	64	2.7	4.8	20	29	19
39	0.31	78	27.5	640	0.12	0.93	59	2.7	4.1	28	28	16
40	0.34	153	20.6	797	0.09	1.03	60	2.9	4.6	28	24	14
41	0.28	59	14.4	1679	0.11	1.08	62	2.4	4.6	26	59	36
42	0.77	72	23.1	528	0.08	0.96	46	2.6	4.8	20	20	9
43	0.26	75	22.1	1224	0.09	1.45	89	3.2	4.0	28	25	22
44	0.31	128	20.2	676	0.08	1.06	54	3.2	4.3	25	17	9
45	0.11	66	14.4	1145	0.10	1.16	48	2.6	4.6	28	48	23
46	0.34	77	23.2	357	0.07	0.97	50	2.1	3.9	19	10	5
47	0.32	68	20.2	608	0.08	1.12	69	2.5	4.9	23	15	10
48	1.04	27	19.9	816	0.04	1.55	56	3.4	4.9	24	19	11

00

STOLONO STOLONL LFCHL NOLITT WT5 ROOTSH PERABUN LAR LEAFNO LEAFL
TOTBIO LITTBIO
01DEE1A 02DEE1B 03DEE1C 04DEE1D 05DEE2A 06DEE2B 07DEE2C 08DEE2D 09DEE3A 10DEE3B
11DEE3C 12DEE3D 13LOM1A 14LOM1B 15LOM1C 16LOM1D 17LOM2A 18LOM2B 19LOM2C 20LOM2D
21LOM3A 22LOM3B 23LOM3C 24LOM3D 25MEN1A 26MEN1B 27MEN1C 28MEN1D 29MEN2A 30MEN2B
31MEN2C 32MEN2D 33MEN3A 34MEN3B 35MEN3C 36MEN3D 37LOW1A 38LOW1B 39LOW1C 40LOW1D
41LOW2A 42LOW2B 43LOW2C 44LOW2D 45LOW3A 46LOW3B 47LOW3C 48LOW3D

Appenix D4

Chemical composition of leaves and roots of *Littorella uniflora* - expressed as % dry weight.

SITE	% NITROGEN	% PHOSPHOROUS
DEE		
SHOOT	2.42	0.20
	2.42	0.21
ROOT	2.26	0.18
	2.18	0.18
LOMOND		
SHOOT	2.65	0.23
	2.72	0.25
ROOT	2.99	0.21
	2.42	0.22
MENTEITH		
SHOOT	3.01	0.25
	3.01	0.25
ROOT	2.67	0.21
	2.70	0.22
LOWES		
SHOOT	2.98	0.29
	3.02	0.29
ROOT	2.50	0.25
	2.51	0.26

Appendix D5

Method used for the Determination of Light Response Curves of Field *Littorella* Samples 1990

Changes in oxygen evolution were carried out using two Rank Oxygen electrodes connected to a dual channel chart recorder. Each electrode was placed in a box constructed of black perspex in which a window was placed in order to admit light. Irradiance levels were altered by placing Balzar neutral density filters in the window. Irradiance was supplied by 24°, 50W dichroic quartz-halogen spot lamps (Wotan). One lamp was used per oxygen electrode.

Tests commenced in the dark in order to obtain the dark respiration rate. The irradiance was then increased in approximately 9 increments to a maximum of 70μmol/m^2/sec. One set of measurements was carried out on the two most recent mature leaves. At the end of each series the dark respiration was again measured.

Calibration of the electrode was carried out by filling the incubation chamber with tap water and leaving until the dissolved gases were in equilibrium with the atmosphere, this was observed as a constant reading on the chart recorder. A few grains of sodium diothionite were then added to the chamber in order to remove any oxygen present in the solution. The deflection recorded for a given change in oxygen concentration could then be calculated. A table of the amount of oxygen dissolved in air equilibrated water at a given temperature can be obtained in Hipkins & Baker (1986).

The chamber was then rinsed repeatedly to remove any trace of diothionite and test media was then added to the chamber and again allowed to equilibrate with the atmosphere.

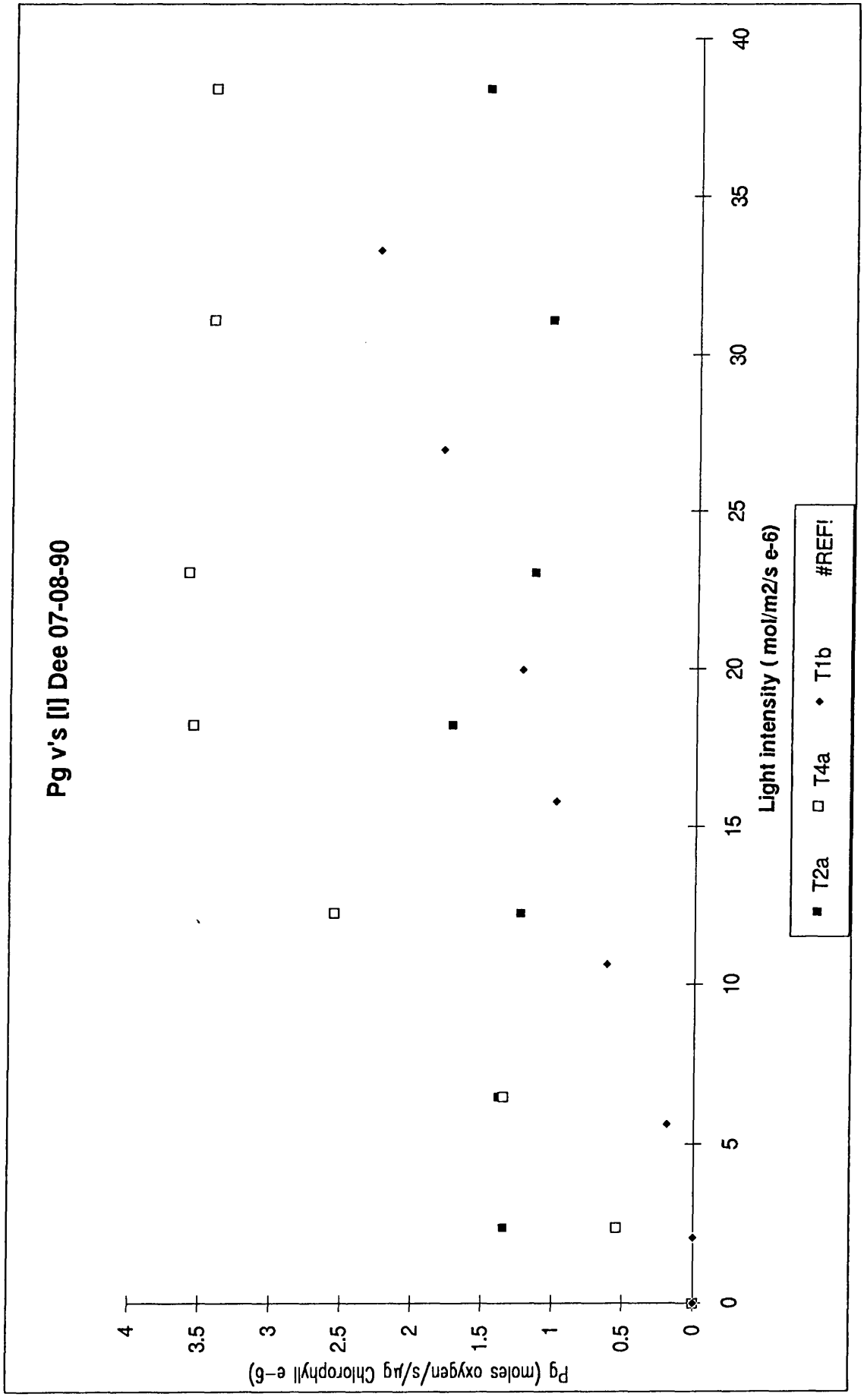
Leaf material was collected from each site as described above and stored in aerated water in natural light until measurements could be carried out. On no occasion did this exceed 48hours. All measurements were carried out at 15 C with a constant stirring rate. Experiments were carried out in distilled water enriched with 1ml per litre of the following stock solution.

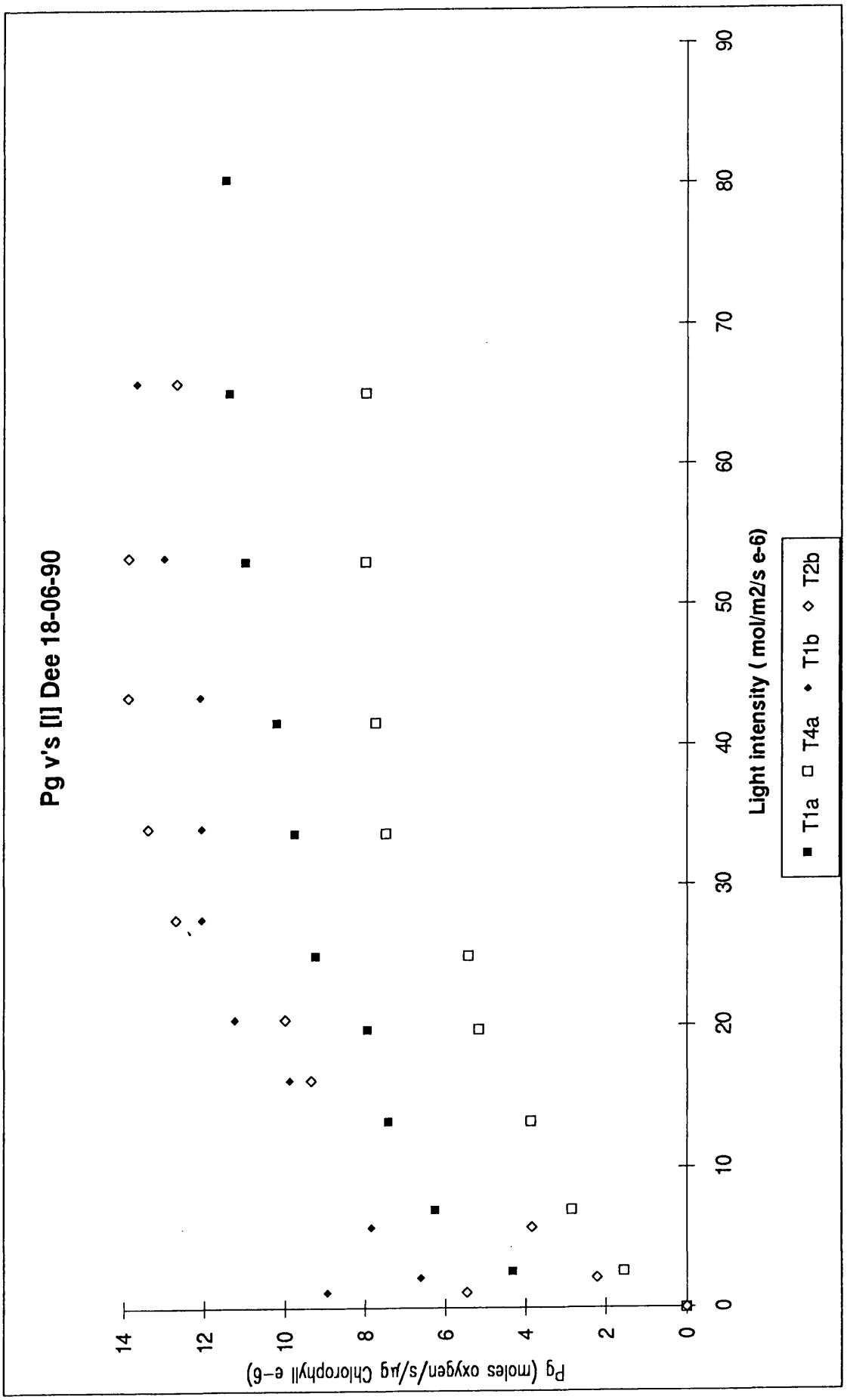
Table D5.1
Stock solution used for light response curve determination

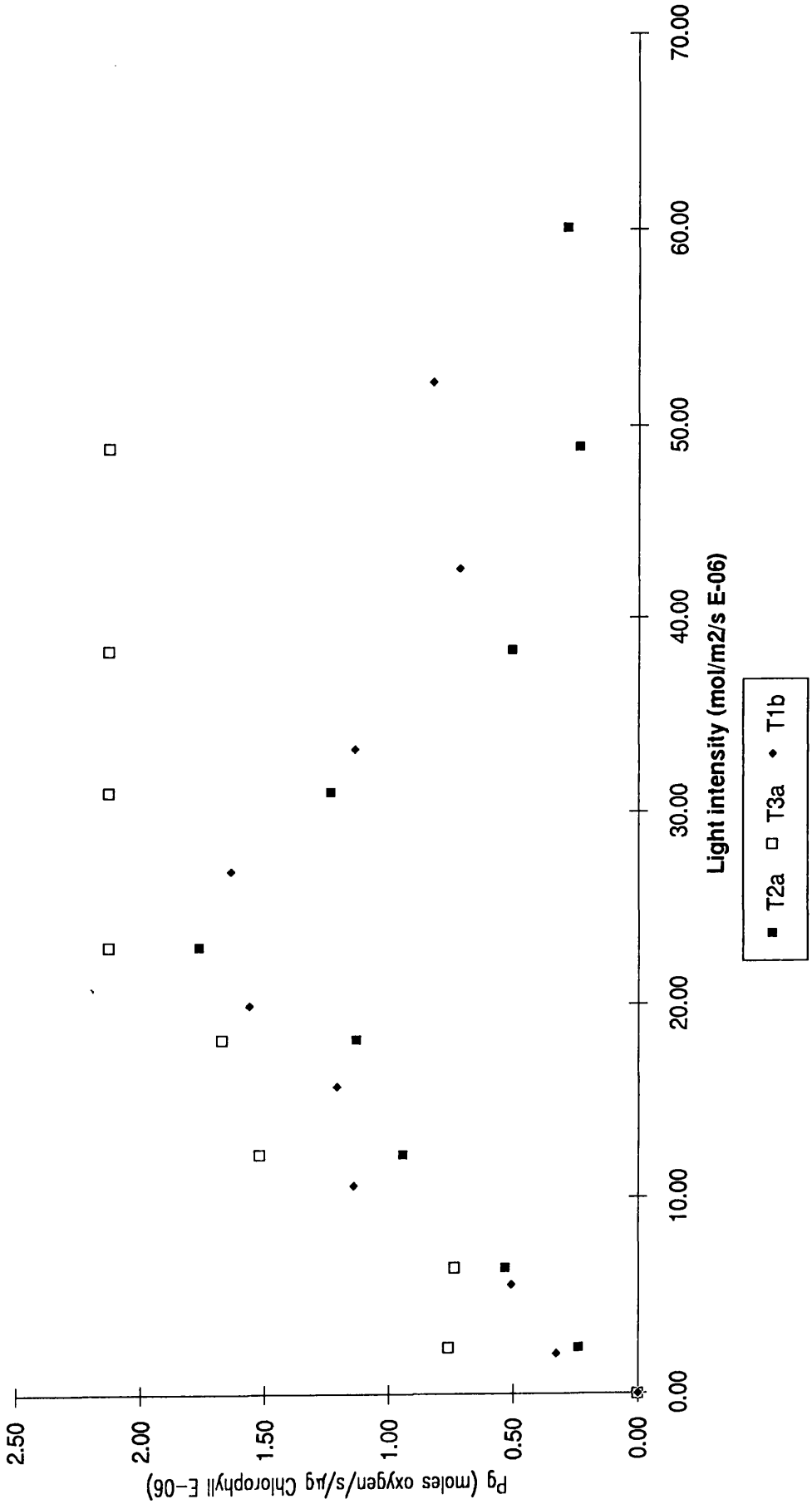
	concentration g/l
$NaNO_3$	1.5
K_2HPO_4	0.04
$MgSO_4$	0.075
$CaCl_2$	0.036
$NaCO_3$	0.83
Adjusted to pH 6.5 using HCl	

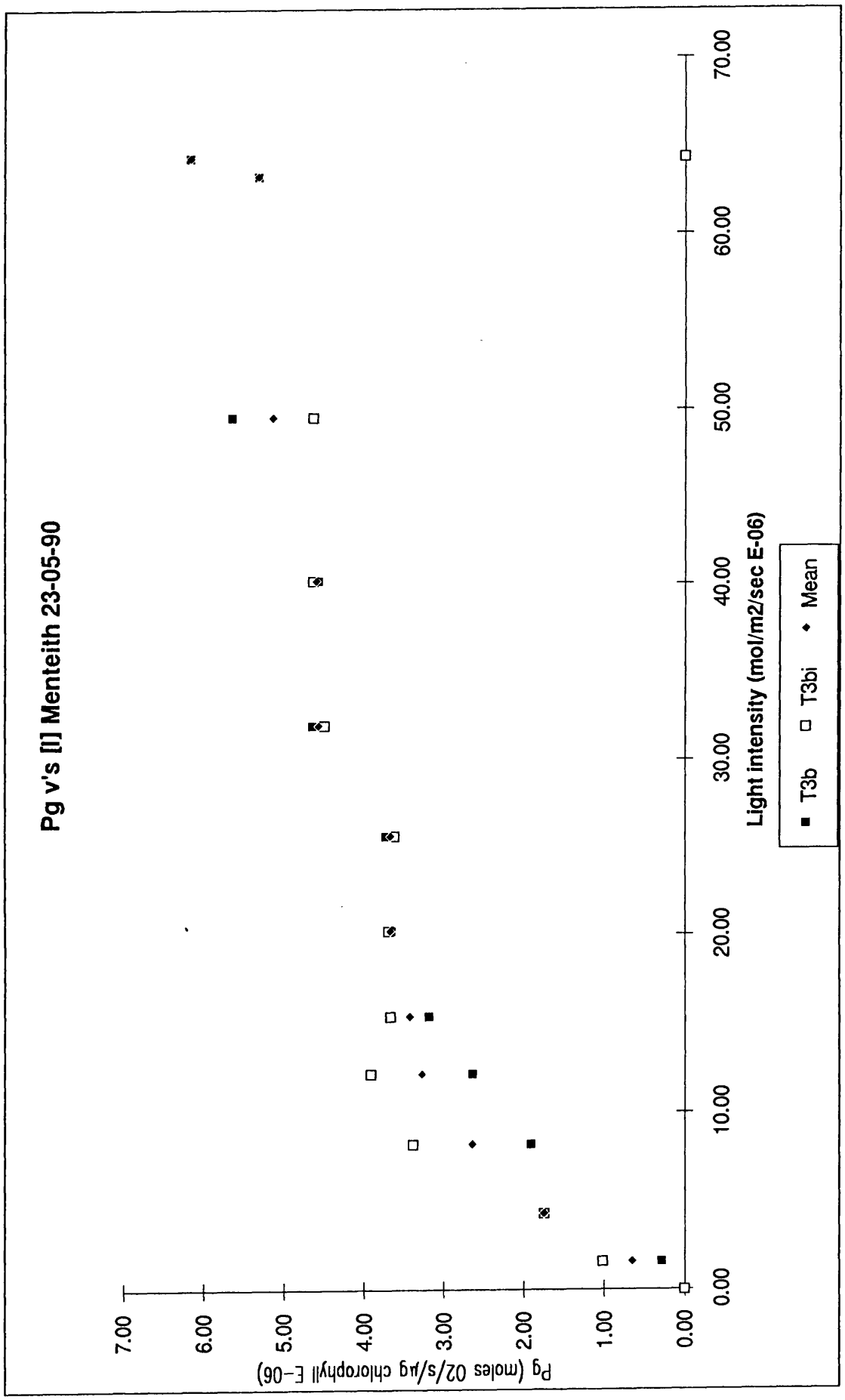
Initially measurements were carried out on leaves horizontally sliced in 1mm sections. However due to the buoyant nature of the leaf sections all the material collected at the top of the incubation chamber, resulting in possible self shading. All subsequent measurements were carried out on two leaves that were sliced vertically and help in position by an 3mm diameter mesh.

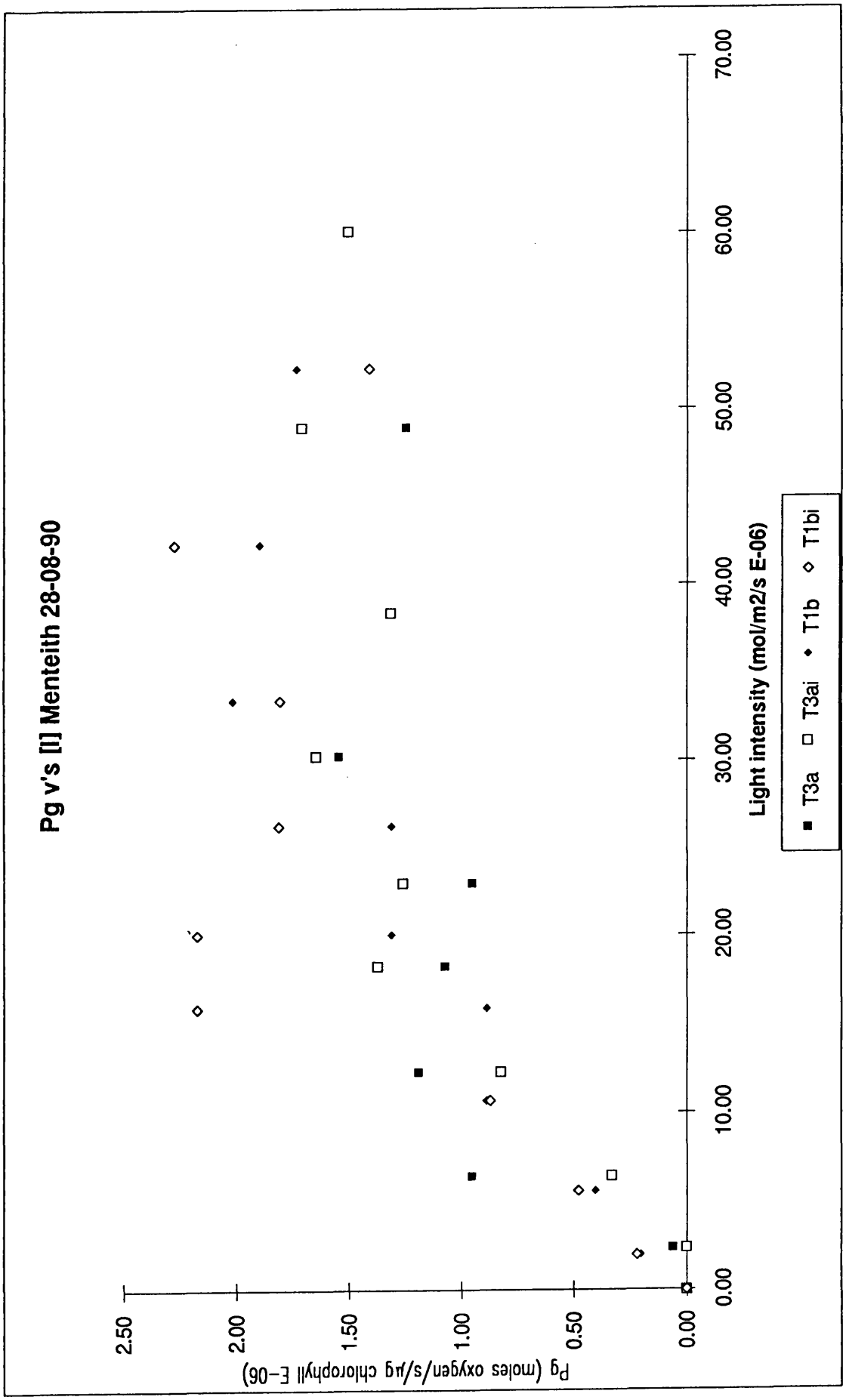
Unfortunately, due to problems with equipment a sufficient number of samples to allow statistical analyses were not obtained. The following plots are of gross photosynthesis per µg chlorophyll, and are included for interest only.

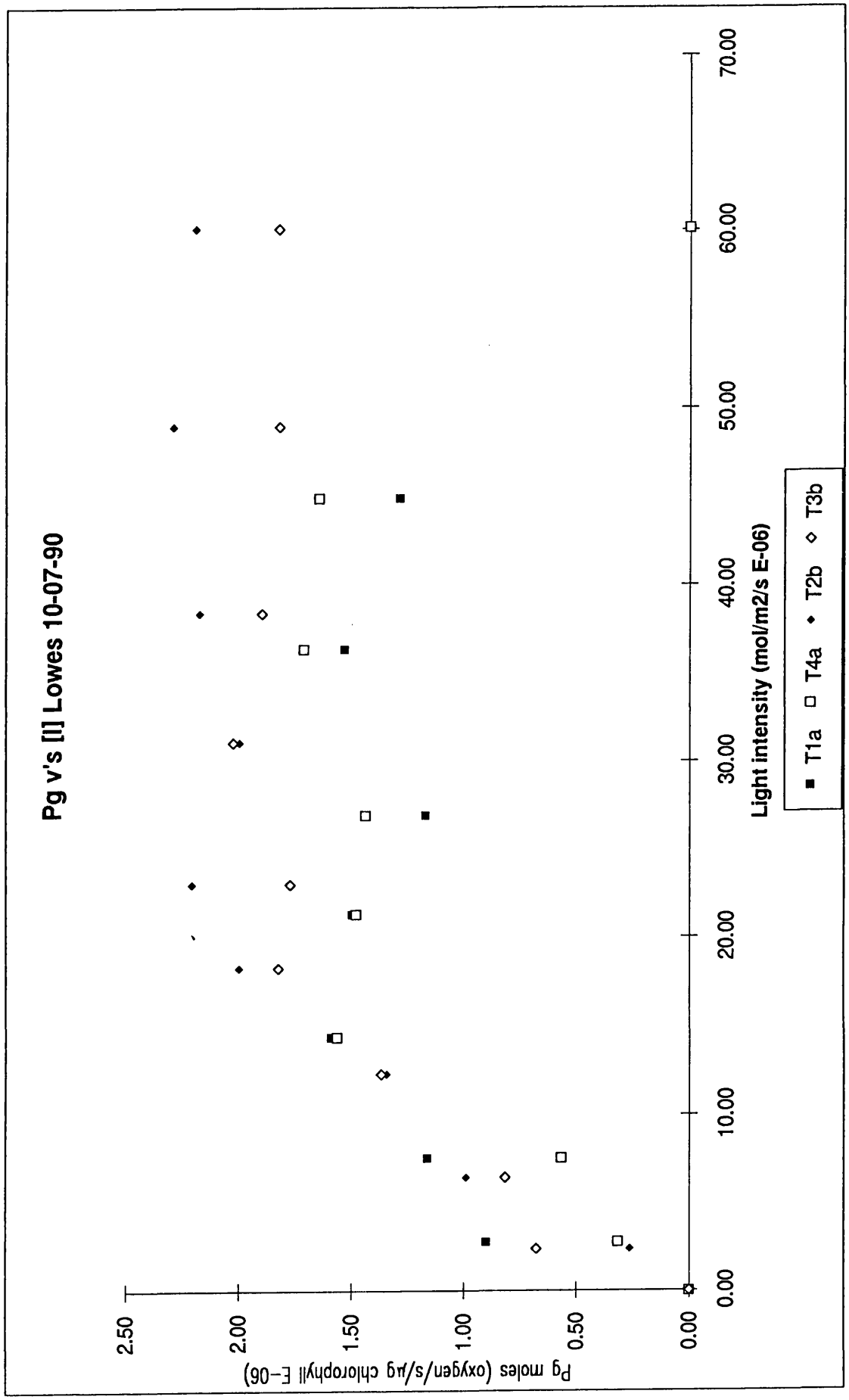

Pg v's [I] Dee 07-08-90
Pg (moles oxygen/s/µg Chlorophyll e-6)
Light intensity (mol/m2/s e-6)
T2a
T4a
T1b
#REF!
0
0.5
1
1.5
2
2.5
3
3.5
4
0
5
10
15
20
25
30
35
40

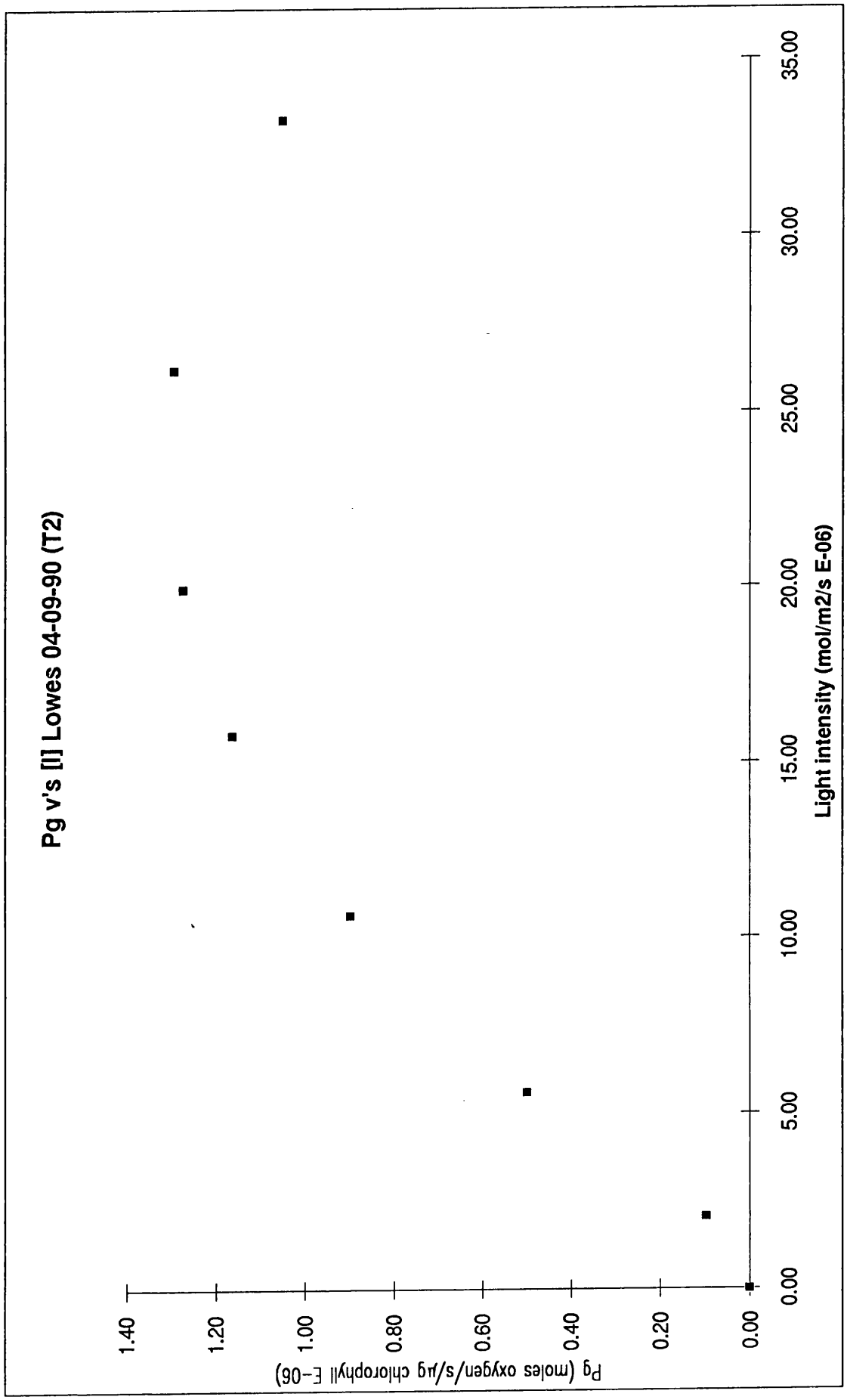

Pg v's [I] Dee 18-06-90
Pg (moles oxygen/s/µg Chlorophyll e-6)
0
2
4
6
8
10
12
14
0
10
20
30
40
50
60
70
80
90
Light intensity (mol/m2/s e-6)
T1a
T4a
T1b
T2b


Pg v's [I] Lomond 23-07-90
Pg (moles oxygen/s/µg Chlorophyll E-06)
2.50
2.00
1.50
1.00
0.50
0.00
0.00
10.00
20.00
30.00
40.00
50.00
60.00
70.00
Light intensity (mol/m2/s E-06)
T2a
T3a
T1b


Pg v's [I] Menteith 23-05-90
Pg (moles 02/s/µg chlorophyll E-06)
7.00
6.00
5.00
4.00
3.00
2.00
1.00
0.00
0.00
10.00
20.00
30.00
40.00
50.00
60.00
70.00
Light intensity (mol/m2/sec E-06)
T3b
T3bi
Mean


Pg v's [I] Menteith 28-08-90
Pg (moles oxygen/s/µg chlorophyll E-06)
2.50
2.00
1.50
1.00
0.50
0.00
0.00
10.00
20.00
30.00
40.00
50.00
60.00
70.00
Light intensity (mol/m2/s E-06)
T3a
T3ai
T1b
T1bi


Pg v's [I] Lowes 10-07-90
Pg moles (oxygen/s/µg chlorophyll E-06)
2.50
2.00
1.50
1.00
0.50
0.00
0.00
10.00
20.00
30.00
40.00
50.00
60.00
70.00
Light intensity (mol/m2/s E-06)
T1a
T4a
T2b
T3b


Pg v's [I] Lowes 04-09-90 (T2)
Pg (moles oxygen/s/µg chlorophyll E-06)
1.40
1.20
1.00
0.80
0.60
0.40
0.20
0.00
0.00
5.00
10.00
15.00
20.00
25.00
30.00
35.00
Light intensity (mol/m2/s E-06)

Appendix E
Results from Greenhouse trials

E.1 Sediment organic content

Table E.1a Biomass Data

Treatment (% organic)	Total biomass of pot (g)				Mean	St.Err
100	0.0461	0.0471	0.0338	0.0395	0.0416	0.0031
75	0.0329	0.1076	0.0753	0.0736	0.0724	0.0153
50	0.0499	0.0973	0.0423	0.0404	0.0575	0.0134
25	0.0526	0.0436	0.0704	0.0208	0.0469	0.0103
10	0.0328	0.0519	0.0230	0.0174	0.0313	0.0076
0	0.0154	0.0178	0.0130	0.0257	0.0180	0.0028

Table E.1b Root biomass

Treatment (% organic)	Weight of roots (g)				Mean	St.Err
100	0.0332	0.0338	0.0231	0.0242	0.0286	0.0029
75	0.0234	0.0195	0.0098	0.0491	0.0255	0.0084
50	0.0382	0.0229	0.0257	0.0250	0.0280	0.0035
25	0.0393	0.0242	0.0378	0.0161	0.0294	0.0056
10	0.0234	0.0371	0.0179	0.0140	0.0231	0.0050
0	0.0097	0.0136	0.0097	0.0201	0.0133	0.0025

Table E1.c Shoot Biomass

Treatment (% organic)	Weight of shoots (g)				Mean	St.Err
100	0.0129	0.0133	0.0107	0.0153	0.0131	0.0009
75	0.0095	0.0154	0.0053	0.0134	0.0109	0.0022
50	0.0117	0.0231	0.0109	0.0153	0.0153	0.0028
25	0.0133	0.0194	0.0104	0.0047	0.0120	0.0031
10	0.0094	0.0148	0.0051	0.0174	0.0117	0.0028
0	0.0057	0.0042	0.0033	0.0056	0.0047	0.0006

Table E.1d Root:Shoot

Treatment (% organic)	Ratio of Root biomass to Shoot biomass				Mean	St.Err
100	2.57	2.54	2.16	1.58	2.21	0.2308
75	2.46	1.27	1.85	3.66	2.31	0.5131
50	3.26	0.99	2.36	1.63	2.06	0.4885
25	2.95	1.25	3.63	3.43	2.82	0.5417
10	2.49	2.51	3.51	0.80	2.33	0.5609
0	1.70	3.24	2.94	3.59	2.87	0.4105

Table E.1e Stolon Biomass

Treatment (% organic)	Weight in g of stolons				Mean	St.Err
100	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000
75	0.0000	0.0237	0.0258	0.0111	0.0152	0.0060
50	0.0000	0.0338	0.0066	0.0000	0.0101	0.0081
25	0.0000	0.0000	0.0222	0.0000	0.0056	0.0056
10	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000
0	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000

Table E.1f Chlorophyll content

Treatment (% organic)	Total chlorophyll (mg/g)				Mean	St.Err
100	12.89	20.53	15.10	16.18	16.18	1.61
75	12.73	12.29	14.57	14.75	13.59	0.63
50	13.59	17.98	13.87	8.99	13.61	1.84
25	11.93	16.44	12.59	23.14	16.03	2.57
10	14.15	13.35	14.51		14.00	0.35
0	26.45	31.75	20.18	20.91	24.82	2.70

Table E.1g Chlorophyll a:b

Treatment (% organic)	Ratio chlorophyll a:b				Mean	St.Err
100	2.95	3.02	3.23	2.72	2.98	0.11
75	3.24	3.00	3.29	3.19	3.18	0.06
50	3.09	3.15	2.75	3.15	3.04	0.10
25	3.06	3.36	2.90	3.40	3.18	0.12
10	3.15	3.29	3.37		3.27	0.06
0	3.26	3.29	3.32	3.14	3.25	0.04

Table E.1h Leaf Area Ratio

Treatment (% organic)	Weight in g of individual plants	Mean	St.Err
100		#DIV/0!	#DIV/0!
75		#DIV/0!	#DIV/0!
50		#DIV/0!	#DIV/0!
25		#DIV/0!	#DIV/0!
10		#DIV/0!	#DIV/0!
0		#DIV/0!	#DIV/0!

Appendix E
Results from Green house trials

E.2 Shading

Table E.2a Biomass Data

Treatment	Total biomass of pot (g)						Mean	St.Err
Shade	0.0150	0.0117	0.0225	0.0075	0.0145	0.0131	0.0141	0.0020
Light	0.0331	0.0151	0.0654	0.0200	0.0374	0.0286	0.0333	0.0072
Shade -Light	0.0382	0.0400	0.0302	0.0102	0.0657	0.0404	0.0375	0.0073
Light only	0.0510	0.0295	0.0743	0.0456	0.0201	0.0530	0.0456	0.0078

Table E.2b Root biomass

Treatment	Weight of roots (g)						Mean	St.Err
Shade	0.0092	0.0105	0.0139	0.0047	0.0098	0.0109	0.0098	0.0012
Light	0.0200	0.0072	0.0273	0.0103	0.0176	0.0094	0.0153	0.0031
Shade -Light	0.0216	0.0171	0.0186	0.0055	0.0289	0.0126	0.0174	0.0032
Light only	0.0219	0.0120	0.0177	0.0191	0.0103	0.0339	0.0192	0.0034

Table E.2c Shoot Biomass

Treatment	Weight of Shoot (g)						Mean	St. Err
Shade	0.0058	0.0012	0.0086	0.0028	0.0047	0.0022	0.0042	0.0011
Light	0.0131	0.0079	0.0381	0.0097	0.0128	0.0125	0.0157	0.0046
Shade -Light	0.0166	0.0110	0.0166	0.0047	0.0262	0.0109	0.0143	0.0030
Light only	0.0291	0.0089	0.0051	0.0111	0.0031	0.0191	0.0127	0.0040

Table E.2d Root:Shoot

Treatment	Ratio of Root biomass to Shoot biomass						Mean	St.Err
Shade	1.59	8.75	1.62	1.68	2.09	4.95	3.45	1.19
Light	1.53	0.91	0.72	1.06	1.38	0.75	1.06	0.14
Shade -Light	1.30	1.55	1.12	1.17	1.10	1.16	1.23	0.07
Light only	0.75	1.35	3.47	1.72	3.32	1.77	2.06	0.45

Table E.2e Stolon Biomass

Treatment	Weight in g of stolons						Mean	St.Err
Shade	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000
Light	0.0000	0.0000	0.0000	0.0000	0.0070	0.0067	0.0023	0.0014
Shade -Light	0.0000	0.0119	0.0000	0.0000	0.0106	0.0169	0.0066	0.0031
Light only	0.0000	0.0086	0.0515	0.0154	0.0067	0.0000	0.0137	0.0079

Table E.2f Chlorophyll content

(% organic)	Total chlorophyll (mg/g)						Mean	St. Err
Shade	44.35	67.67	36.29	55.67			51.00	6.83
Light	13.59	17.98	13.87	8.99			13.61	1.84
Shade -Light	16.18	18.83	11.92	20.27	13.89	21.30	17.07	1.51
Light only	17.25	22.39	15.89	16.72	22.12	14.52	18.15	1.35

Table E.2g Chlorophyll a:b

(% organic)	Ratio chlorophyll a:b						Mean	St. Err
Shade	2.51	2.38	2.31	2.72			2.48	0.09
Light	3.09	3.15	2.77	3.15			3.04	0.09
Shade -Light	2.79	2.81	2.64	2.61	2.51	2.57	2.66	0.05
Light only	2.64	2.81	2.64	2.51	2.84	2.51	2.66	0.06

Table E.1h Leaf Area Ratio

(% organic)	Mean	St. Err
Shade	#DIV/0!	#DIV/0!
Light	#DIV/0!	#DIV/0!
Shade -Light	#DIV/0!	#DIV/0!
Light only	#DIV/0!	#DIV/0!

Appendix E.2 Raw Data From Long Term Greenhouse Shading Experiment

Unshaded plants (1)

Plant no.	initial no. leaves 17/2/92	30/3/92	net change 30/3/92	13/4/92	net change 13/4/92
04	4	6	(+)2	5	(-)1
14	3	4	(+)1	5	(+)1
16	5	7	(+)2	8	(+)1
10	3	4	(+)1	6	(+)2
02	3	4 + 1y + 1s	(+)1 (+)1st	6 + st + pt	(+)1 (+)1pt
18	3	5 + 1st	(+)2 (+)1st	5 + st + pt	(+)1pt
Total	21	30 + 2st	(+)9 (+)2st	35 + 2st +2pt	(+)4 (+)2pt

Key:
st - stolon
pt - plant
y - yellow leave
(+) - increase
(-) - decrease

Shaded Plants

Plant no.	initial no. leaves 17/2/92	30/3/92	net change 30/3/92	13/4/92	net change 13/4/92
23	3	3 + 1y	0	4	(+)1
35	2	2 + 1y	0	2	0
09	4	3 + 1y	(-)1	3	0
20	4	4	0	3	(-)1
30	3	3	0	4	(+)1
32	3	3	0	2	(-)1
Total	19	18	(-)1	18	0

Unshaded Plants (2)

Plant no.	initial no. leaves 17/2/92	30/3/92	net change 30/3/92	4/5/92	net change 4/5/92	13/8/92	net change 13/8/92
15	3	4	(+)1	4 + 1y	0	7	(+)3
17	3	5	(+)2	4 + 1st	(-)1 (+)1st	4 + st + pt	0 (+)1pt
22	3	5	(+)2 (+)1st	5 + st + 2pt	0 (+)2pt	4 + st + 4pt	(-)1 (+)4pt
27	3	5 + 1st	(+)1	3 + 1y	(-)1	5 + st + 2pt	(+)2 (+)st (+)2pt
29	2	4	(+)2	3	(-)1	4 + st + pt	(-)1 (+)st (+)pt
36	3	4 + 1y	(+)2	4 + 1st	(-)1 (+)1st	6	(+)2 (-)st
Total	17	27 + 1st	(+)10 (+)1st	23 +3st + 2pt	(-)4 (+)2st (+)2pt	26 + 4st + 8pt	(+)5 (-)st (+)7pt

Shaded then unshaded plants

Plant no.	initial no. leaves 17/2/92	30/3/92	net change 30/3/92	13/4/92	net change 13/4/92	13/8/92	net change 13/8/92
23	3	3 + 1y	0	4	(+)1	9	(+)5
35	2	2 + 1y	0	2	0	6 + 2st + 4pt	(+)1 (+)2st (+)4pt
09	4	3 + 1y	(-)1	3	0	5	(+)3
20	4	4	0	3	(-)1	5	(+)1
30	3	3	0	4	(+)1	9 + 3st + 4pt	(+)7 (+)3st (+)4pt
32	3	3	0	2	(-)1	7 + st + 6pt	(+)3 (+)st (+)6pt
Total	19	18	(-)1	18	0	41 + 6st+ 14pt	(+)20 (+)6st (+)14pt

Appendix F.1
Change in chlorophyll concentration in leaves of *Littorella* after the application of shading

Treatment	Day	Total Chlorophyll (mg/g dry weight)				mean (se)
Control	1	15.86	17.30	19.85	14.88	16.97(1.08)
Control	3	11.30	16.87	17.25	11.03	14.11(1.70)
Control	6	13.67	14.77	16.92	15.82	15.29(0.70)
Control	10	11.95	15.92	18.66	15.18	15.43(1.38)
Control	12	12.80	15.98	15.67	14.90	14.83(0.72)
Shade	1	15.86	17.30	19.85	14.88	16.97(1.08)
Shade	2	12.30	13.46	14.20	13.84	13.45(0.41)
Shade	3	15.09	13.77	13.91	14.28	14.26(0.30)
Shade	4	14.22	22.48	13.52	13.10	15.83(2.23)
Shade	6	19.00	17.13	16.60	*	17.57(0.73)
Shade	9	17.49	16.64	16.11	16.59	16.59(0.29)
Shade	10	22.42	19.58	20.02	*	20.67(0.88)
Shade	12	18.14	21.53	22.02	21.79	20.87(0.92)

Appendix F2
Gross Photosynthesis - measured and derived values

Control			White Shade			Ulva Shade				
[I]	Pg	Pg	[I]	Pg	Pg	[I]	Pg	Pg		
	measured	derived		measured	derived		measured	derived		
0.0	0.000	0.000	0.0	0.000	0.000	0.0	0.000	0.000	0.000	0.000
14.1	0.177	0.111	14.1	0.137	0.246	6.2	0.109	0.108	0.000	0.099
30.0	0.212	0.206	30.0	0.326	0.343	13.9	0.181	0.190	0.000	0.188
47.0	0.285	0.284	47.0	0.397	0.394	25.5	0.253	0.264	0.013	0.280
77.8	0.285	0.385	77.8	0.456	0.438	36.5	0.325	0.307	0.258	0.340
95.6	0.520	0.429	95.6	0.567	0.453	47.7	0.325	0.337	0.345	0.386
143.0	0.585	0.513	143.0	0.502	0.476	74.2	0.434	0.381	0.519	0.456
194.0	0.630	0.572	194.0	0.534	0.489	101.0	0.434	0.405	0.563	0.500
210.0	0.630	0.587	210.0	0.471	0.492	112.0	0.434	0.413	0.563	0.513
260.0	0.585	0.623	260.0	0.426	0.499	136.0	0.434	0.425	0.563	0.536
331.0	0.630	0.661	331.0	0.567	0.505	177.0	0.434	0.440	0.563	0.564
396.0	0.563	0.686	396.0	0.486	0.509	212.0	0.434	0.448	0.519	0.580
525.0	0.541	0.719	525.0	0.456	0.514	298.0	0.434	0.461	0.563	0.606
						330.0	0.343	0.464	0.563	0.612
0.0	0.000	0.000	0.0	0.000	0.000					
11.5	0.000	0.137	11.7	0.139	0.213	0.0	0.000	0.000	0.000	0.000
28.7	0.134	0.269	21.8	0.296	0.295	11.7	0.071	0.106	0.243	0.315
42.3	0.214	0.339	43.2	0.341	0.378	21.8	0.160	0.164	0.379	0.419
65.5	0.409	0.422	71.2	0.436	0.426	43.2	0.233	0.238	0.339	0.516
81.1	0.467	0.461	89.3	0.541	0.444	71.2	0.326	0.290	0.434	0.569
118.0	0.555	0.525	128.0	0.523	0.467	89.3	0.345	0.312	0.574	0.588
147.0	0.555	0.559	172.0	0.523	0.481	128.0	0.365	0.342	0.574	0.612
190.0	0.648	0.594	180.0	0.560	0.483	172.0	0.384	0.363	0.517	0.627
233.0	0.617	0.619	228.0	0.505	0.492	180.0	0.384	0.365	0.517	0.629
298.0	0.586	0.644	294.0	0.487	0.500	228.0	0.384	0.379	0.631	0.647
			346.0	0.487	0.505	294.0	0.384	0.391	0.661	0.651
0.0	0.000	0.000	456.0	0.470	0.511	346.0	0.365	0.398	0.720	0.657
11.7	0.068	0.172	537.0	0.420	0.513	456.0	0.365	0.407	0.720	0.660
21.8	0.248	0.261				537.0	0.365	0.454	0.631	
43.2	0.339	0.371	0.0	0.000	0.000					
71.2	0.480	0.447	11.7	0.102	0.145	0.0	0.000	0.000		
89.3	0.529	0.478	21.8	0.190	0.192	14.1	0.065	0.163		
128.0	0.579	0.520	43.2	0.212	0.237	30.0	0.236	0.259		
172.0	0.579	0.548	71.2	0.261	0.261	47.0	0.323	0.319		
180.0	0.579	0.552	89.2	0.286	0.269	77.8	0.504	0.403		
228.0	0.579	0.571	128.0	0.295	0.280	95.6	0.481	0.440		
294.0	0.579	0.587	172.0	0.295	0.287	143.0	0.504	0.462		
346.0	0.554	0.596	180.0	0.295	0.288	194.0	0.481	0.467		
456.0	0.554	0.609	228.0	0.295	0.292	210.0	0.471	0.480		
537.0	0.554	0.615	294.0	0.295	0.296	260.0	0.481	0.492		
			246.0	0.286	0.293	331.0	0.504	0.499		
0.0	0.000	0.000	455.0	0.286	0.300	396.0	0.481	0.508		
11.7	0.156	0.163	536.0		0.302	525.0	0.435			
21.8	0.288	0.249								
43.2	0.357	0.356				0.0	0.000	0.000		
71.2	0.444	0.431				11.5	0.141	0.150		
89.2	0.480	0.461				28.7	0.337	0.277		
128.0	0.468	0.503				42.3	0.410	0.339		
172.0	0.468	0.532				65.5	0.471	0.407		
180.0	0.470	0.536				81.1	0.586	0.438		
228.0	0.421	0.554				118.0	0.519	0.486		
294.0	0.580	0.571				147.0	0.552	0.510		
346.0	0.550	0.580				190.0	0.487	0.535		
445.0	0.499	0.592				233.0	0.440	0.522		
536.0	0.501	0.599				298.0	0.586	0.569		
						351.0	0.503	0.579		
						459.0	0.471	0.592		
						545.0		0.622		

Appendix F.3

$\Delta^{13}C$ values, % nitrogen and % carbon (dry weight) in the leaves of *Littorella* three weeks after the application of shading.

Treatment	%N	%C	ΔPBD
Control	1.80 1.83	40.92 40.57	-26.10 -26.61
	2.04 2.05	36.97 37.51	-27.02 -27.27
	2.19 2.22	39.37 39.12	-24.45 -25.45
means(se)	2.02(0.03)	40.43(0.33)	-26.15(0.43)
Ulva	2.69 2.54	43.51 38.67	-30.21 -29.40
	2.79 2.79	37.80 38.14	-29.73 -29.63
	2.93 2.93	37.87 36.91	-29.38 -29.10
means(se)	2.71(0.04)	38.82(0.97)	-29.58(0.15)
white	2.64 2.56	40.64 40.31	-30.00 -30.18
	2.68 2.71	39.54 41.63	-28.82 -29.57
	2.75 2.72	40.94 39.54	-28.70 -28.42
means(se)	2.68(0.03)	40.43(0.33)	-29.28(0.30)

Appendix F.4

Chlorophyll concentration in the leaves of *Littorella* three weeks after the application of shading

Treatment	Total Chlorophyll (mg/g dry weight)	Chlorophyll *a*:*b*
Control	12.07 14.15 14.51	2.49 2.63 2.81
	15.63 15.98 14.73	2.84 2.74 3.00
mean(se)	14.51(0.56)	
Ulva shade	19.65 18.96 21.74	2.62 2.71 2.73
	18.86 24.24 17.07	2.75 2.61 2.52
mean(se)	20.09(1.03)	
White shade	19.95 17.62 19.68	2.64 2.69 2.28
	17.74 13.33 16.29	2.53 2.50 2.55
mean(se)	17.44(0.99)	

ppendix G: **Publication**

ARRS, S.J., MURPHY, K.J., HILLS, J.M. (1993): The aquatic lora of Loch Dee, 1904-1990. In: Proceedings of the Loch Dee mposium. Acidification, Forestry and Fisheries in Upland alloway. Foundation for Water Research, 97-106.

PROCEEDINGS OF THE LOCH DEE SYMPOSIUM

Acidification, forestry and fisheries management in upland Galloway

Held at the Cally Palace Hotel, Gatehouse of Fleet,
Dumfries and Galloway, Scotland
8 - 9 December 1992

Published by

FWR FOUNDATION *for* WATER RESEARCH

PROCEEDINGS

OF THE LOCH DEE SYMPOSIUM

ch Dee is one of a series of lochs situated in the Southern Uplands of south west Scotland where oblems of declining fish stocks linked to increasing acidification were first noted in Britain. In the 70's several lochs and upland parts of large riverine systems suffered falling or complete loss of lmonid populations.

e **Loch Dee Project** was set up in 1980. Loch Dee showed a historical decline in pH from before turn of the century, but still had a brown trout population. Approximately 30% of the Loch chment was planted with conifers in 1973 - 1975. A major hydrochemical, geochemical and logical study is being undertaken at the site to investigate the effects of acidic deposition and estry plantations on the catchment, streamwater and loch ecology.

teering group comprising the Solway River Purification Board, The Scottish Office Environment partment and Agriculture and Fisheries Departments, Forest Enterprise and The Forest Authority iew progress of the project. Many other research organisations are also involved in the study.

e **Symposium programme** extended over two days. Invited speakers reviewed the work lertaken at the site. A wide range of topics was covered including basic chemical and hydrological ormation to studies of the complex interactions of soils, soil water and streamwater. A number poster presentations were also on display during the meeting.

A two day Symposium held at the Cally Palace Hotel,
Gatehouse of Fleet, Dumfries and Galloway, Scotland,
on 8 - 9 December 1992

THE AQUATIC FLORA OF LOCH DEE 1904-1990

*S.J. Marrs, K.J. Murphy, J.M. Hills

Department of Botany, University of Glasgow, Glasgow. Scotland G12 8QQ. *Address for correspondence: Glasgow Marine Technology Centre, James Watt Building, University of Glasgow, Glasgow, Scotland. G12 8QQ.

ABSTRACT

The aquatic flora of Loch Dee (Lat. 55°5'N, Long 4°24'W) is currently dominated by macrophytes of the isoetid growth form, such as *Isoetes lacustris* L., *Lobelia dortmanna* L. and *Littorella uniflora* (L.) Asch. Widespread growth of *Sphagnum* spp., a feature commonly associated with lochs experiencing water deterioration due to acidification, was not observed in any of the three surveys carried out during the last decade. However, work carried out in 1986 showed that *Juncus bulbosus* Schultz, an acid-tolerant species, was abundant in the loch.

A survey in 1990 noted the widespread distribution of *Utricularia* c.f. *stygia* Thor at depths greater than 1m. The last record of *Utricularia* in Loch Dee was that of West in 1904/5 where it is recorded as *Utricularia intermedia* Hayne. The increase of *Utricularia* after liming treatments has been observed in other lochs experiencing acidification, e.g. Loch Fleet (Battarbee, Logan, Murphy, Raven, Aston, & Foster, 1992).

Principle Components Analysis (PCA) of the submerged macrophyte species recorded from four surveys conducted between 1904 - 1990 showed that the loch community in 1990 was more similar to the flora in 1904 than in either 1983 or 1986. This similarity of the loch aquatic flora between 1904 and 1990 was primarily due to the re-occurrence of *Utricularia.*

INTRODUCTION

One of the commonest ecological groups of aquatic macrophytes in Scotland is the isoetids (Farmer and Spence, 1986). These plants are characterised by having short stems; stiff green leaves; a rosette growth form, and high percentage of the internal volume being taken up by gas-filled lacunae (Den Hartog & Segal, 1964). A decline of these species has been observed in many northern temperate areas that have been affected by acidic deposition, for example: Britain (Farmer, 1990); The Netherlands (Arts, de Haan, Siebum & Verheggen, 1989; Morris, 1991) and Sweden (Grahn, 1985).

In certain areas of Scotland, oligotrophic, low pH waterbodies dominated by isoetids, of which Loch Dee is a good example, are prone to acidification (Last, 1989). Relatively little attention has been paid to the effects of acidification on the aquatic macrophyte communities of lochs in Scotland. In this paper we examine the evidence for changes in aquatic flora in relation to both acidification and liming programmes to ameliorate acidification in Loch Dee.

THE SURVEYS

There have been four surveys of the aquatic flora of Loch Dee since 1904. The first survey was carried out in 1904/5 by G. West. This formed part of an extensive survey of Scottish lochs in which 72 Galloway lochs as well as lochs from other areas of Scotland were surveyed over a period of two years, with work being carried out regardless of season or weather conditions (West, 1910). The survey technique consisted of a shoreline survey only. There is no indication of the time of year when the survey was carried out. However, many species inhabiting Loch Dee are perennial and remain identifiable throughout the year. West consulted specialists for identification of species, and although some individual species may have been missed, the identification of those found is likely to be accurate. Species were recorded on a presence/absence basis.

The next recorded survey was not carried out until 1983 as part of an assessment of water quality in Galloway lochs (Raven, 1985). Although this survey was also shore-based, good water clarity allowed reasonable determination of species in the littoral zone. Only 25% of the shoreline was surveyed which may have resulted in some less common species being missed. Data from the survey (carried out in July 1983) were presented on a subjective scale with plants being recorded as present (rare), locally frequent and abundant.

The most detailed survey was that of Murphy, Miller and Anderson (1986). Survey work carried out in June-August 1986 used a variety of techniques including shoreline surveys, transect surveys, frequency sampling using a quadrat, an Ekman grab (Ekman, 1911) and in the deeper water quadrats were placed with the aid of SCUBA. In shallow and deep water standing crop samples were collected using a Lambourn sampler (Hiley, Wright & Berrie, 1981) and an Ekman grab respectively. Species distribution maps were constructed using data from all the above sampling techniques. The location of sample sites visited in 1986 are depicted in Figure 1.

The most recent work on the aquatic flora was carried out during 1990 and 1991 as part of a study in macrophyte/algal relationships in four lochs of differing nutrient status in Scotland (Marrs, Murphy & Dominy, 1993). The study was concentrated in two areas of the loch with one site being located on the south-west shore and two sites in the north-east basin. During 1991 filamentous algal biomass estimates were carried out from April to September by removing the biomass contained in a 17cm diameter collar with a small sieve. Dry weight determinations of filamentous algal biomass were by oven drying samples at 90 C until a constant weight was obtained. In September 1990 a survey of the species in the south-west basin was carried out by two SCUBA divers with species being recorded on a presence/absence basis.

STATISTICAL ANALYSIS

An ordination method appropriate to the analysis of a plant community data set lacking environmental data is Principal Components Analysis (PCA) (Gauch, 1982). PCA was carried out on survey data of submerged aquatic plants at Loch Dee using the CANOCO package (Ter Braak, 1988). The data set consisted of presence/absence

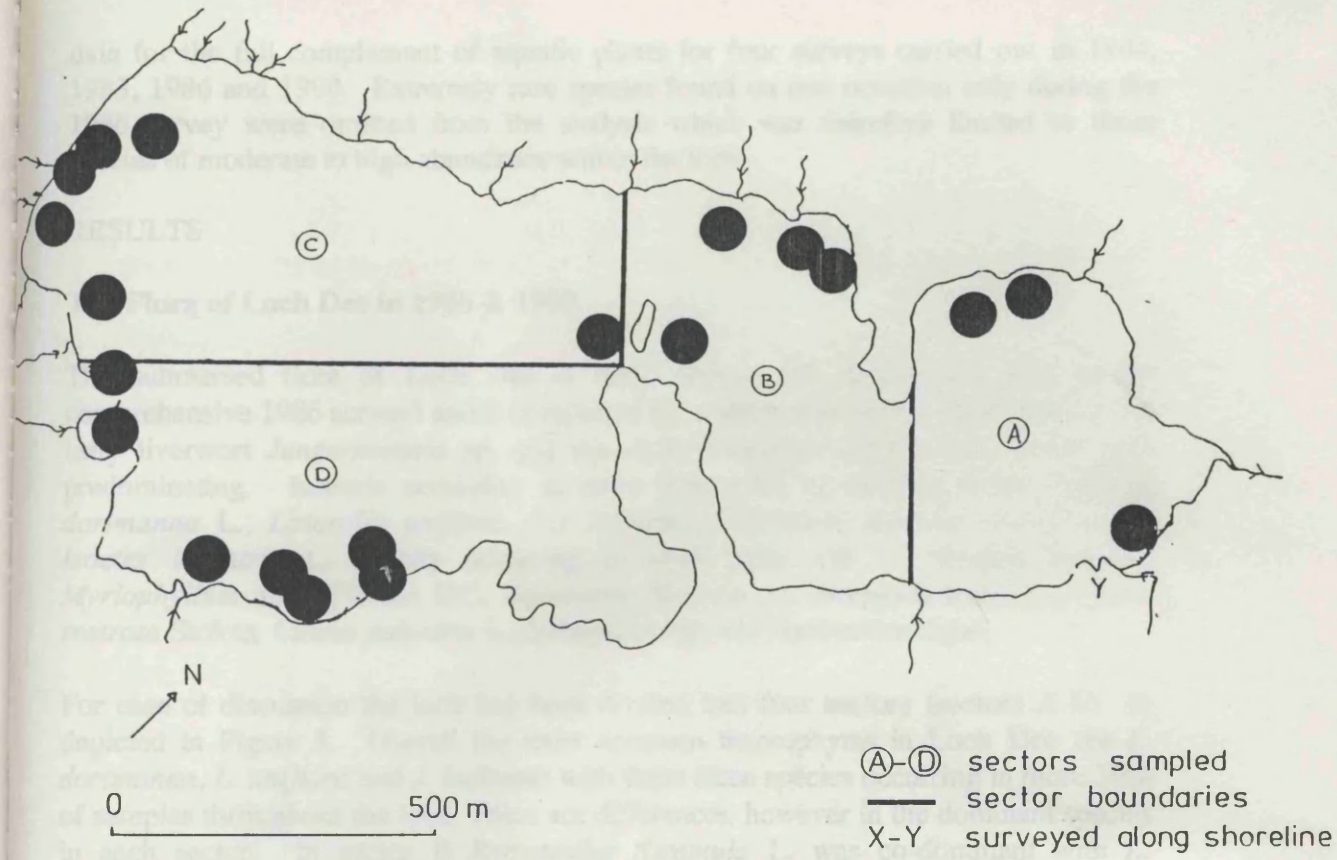


Figure 1. Location of sample sites visited by Murphy *et al.*, 1986.

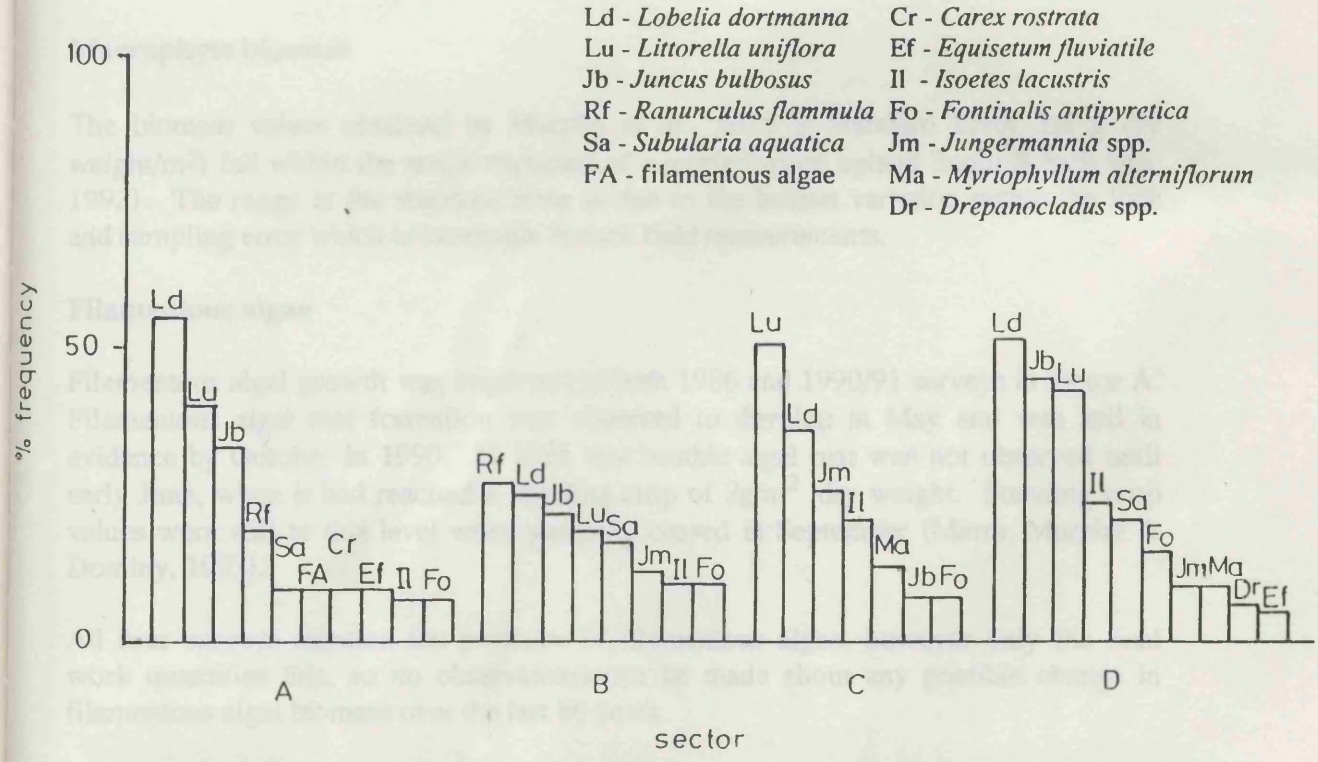


Figure 2. Relative abundance (as % frequency of occurrence in vegetation samples from sector) of dominant macrophyte taxa in sectors A - D. From Murphy *et al.*, 1986.

data for the full complement of aquatic plants for four surveys carried out in 1904, 1983, 1986 and 1990. Extremely rare species found on one occasion only during the 1986 survey were omitted from the analysis which was therefore limited to those species of moderate to high abundance within the loch.

RESULTS

The Flora of Loch Dee in 1986 & 1990

The submersed flora of Loch Dee is fairly diverse (30 species recorded in the comprehensive 1986 survey) and is dominated by isoetids with *Juncus bulbosus* L., the leafy liverwort *Jungermannia* sp. and the moss *Fontinalis antipyretica* Hedw. also predominating. Isoetids occurring in more than 10% of samples were *Lobelia dortmanna* L., *Littorella uniflora* (L.) Acherson, *Subularia aquatica* Schrank and *Isoetes lacustris* L. Taxa occurring in more than 1% of samples included *Myriophyllum alterniflorum* DC., *Equisetum fluviatile* L., *Drepanocladus* sp., *Carex rostrata* Stokes, *Caltha palustris* L., *Sphagnum* spp and filamentous algae.

For ease of discussion the loch has been divided into four sectors (sectors A-D) as depicted in Figure 1. Overall the most common macrophytes in Loch Dee are *L. dortmanna*, *L. uniflora* and *J. bulbosus* with these three species occurring in more 30% of samples throughout the loch. There are differences, however in the dominant species in each sector. In sector B *Ranunculus flammula* L. was co-dominant with *L. dortmanna*. In sectors A and D the latter species was dominant, but this was replaced by *L. uniflora* as the most common species in sector C. *J. bulbosus*, one of the three most common species in sectors A, B & D, was less common in sector C, being replaced by the liverwort *Jungermannia* sp. The abundance of species (as % frequency) in each of the four sectors is summarised in Figure 2.

Macrophyte biomass

The biomass values obtained by Murphy *et al.*, (20.9 ± Standard Error 3.2 g dry weight/m^2) fall within the range expected of a nutrient-poor upland Scottish loch (Ali, 1992). The range in the standard error is due to the habitat variation within the loch and sampling error which is inevitable in such field measurements.

Filamentous algae

Filamentous algal growth was important in both 1986 and 1990/91 surveys in sector A. Filamentous algal mat formation was observed to develop in May and was still in evidence by October in 1990. In 1991 this benthic algal mat was not observed until early June, when it had reached a standing crop of $2g/m^2$ dry weight. Standing crop values were still at this level when sampling ceased in September (Marrs, Murphy & Dominy, 1993).

All four surveys mention the presence of filamentous algae, however only the final work quantifies this, so no observations can be made about any possible change in filamentous algal biomass over the last 86 years.

TABLE 1

Species recorded 1904-1990

	West 1904-5	Raven 1983	Murphy *et al.* 1986	Marrs 1990
Isoetids				
Lobelia dortmanna	+	3	3	3
Littorella uniflora	+	1	3	3
Isoetes lacustris	+	3	2	3
Isoetes echinospora			1	
Subularia aquatica	+	1	2	2
Other submersed macrophytes				
Juncus bulbosus	+	2	3	2
Eleocharis multicaulis		1		
Potamogeton polygonifolious	+		1	1
Eleogiton fluitans	+			
Myriophyllum alterniflorum	+	1	2	1
Elatine hexandra	+	1	2	1
Callitriche hamulata	+	1	1	1
Utricularia intermedia (⇒ *U.* c.f. *stygia* 1990)	+			2
Utricularia vulgaris	+			
*Filamentous algae	+	+	1	2
Floating foliage				
Potamogeton natans		1	1	+
Glyceria fluitans			1	
Spaganium augustifolium	+	1	1	
Lemna minor			1	
Shallow water plants				
Rannunculus flammula			2	2
Montia fontana			1	
Emergent species				
Menyanthes trifoliata			1	+
Eleocharis palustris	+	1	1	+
Equisetum fluviatile	+	1	2	+
Carex rostrata	+	2	1	+
Carex lasiocarpa	+	1	1	+
Hydrocotyle vulgaris			1	+
**Phragmites australis*			1	
Bryophytes				
**Fontinalis antipyretica*		1	2	1
**Sphagnum* spp.			2	1
**Drepanocladus* sp.		1		
**Rhizomnium* sp.			1	
**Rynchostegium riparioids*			1	
**Polytrichum commune*			1	
**Pellia* sp.	+		1	
**Jungermannia* sp.	+		2	

West: no abundance data (+ = present)

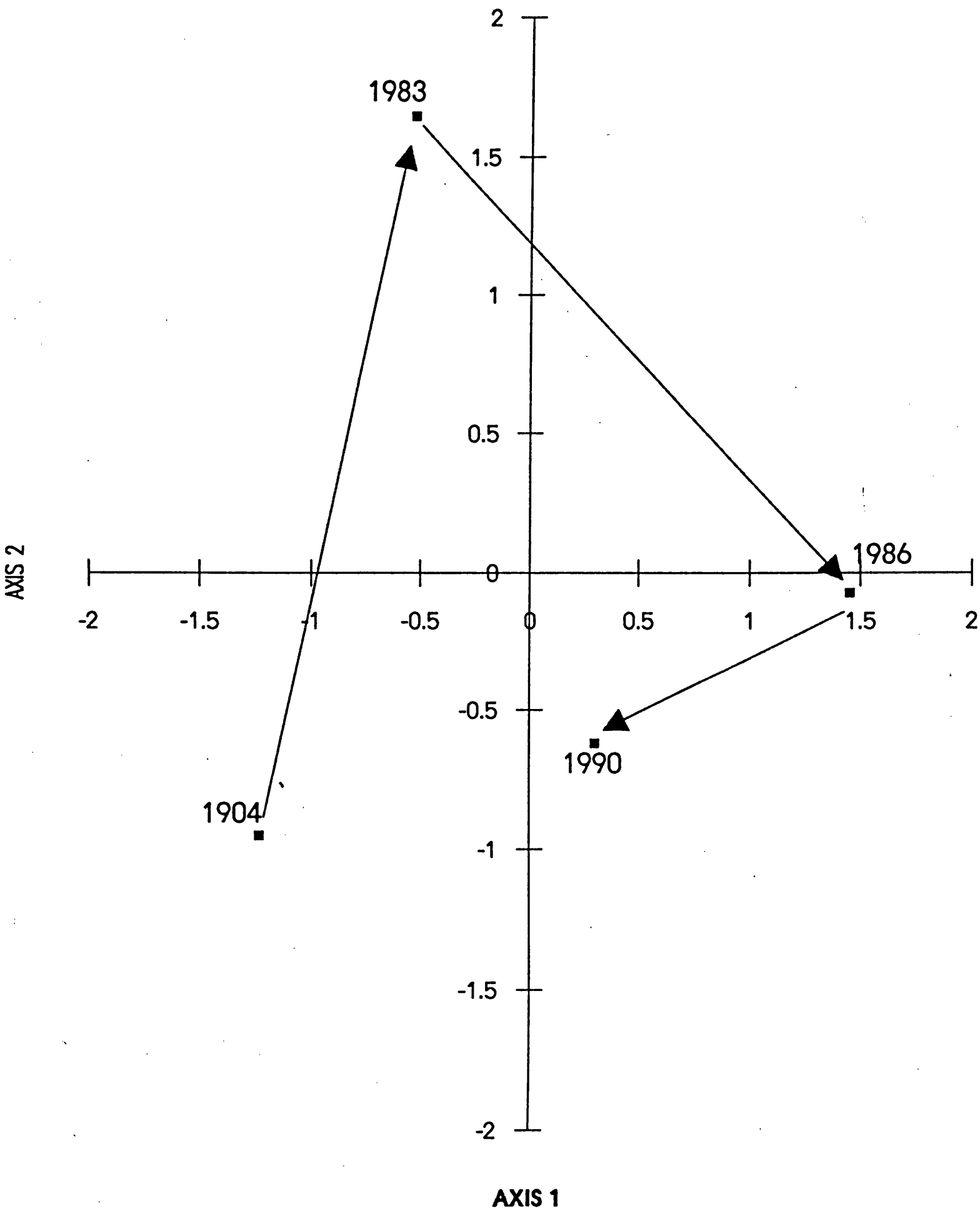
Raven & Marrs: 1 = rare; 2 = locally frequent; 3 = abundant

Marrs, emergent species only: no abundance data (+ = present)

Murphy *et al.*: 1 = present ≤ 5%; 2 = 5-29%; 3 = ≥ 30% (% frequency)

* not included in PCA

Fig. 3. Principal Components Analysis of aquatic plant surveys of Loch Dee; 1904, 1983, 1986 and 1990 (eigenvalue: axis 1 = 0.59, axis 2 =0.23).



Statistical analysis

The PCA was an effective method for summarising and elucidating differences in the aquatic flora of Loch Dee from the period 1904 to 1990. The first axis explained 59% of the variation in the species abundance data set; much of the variation in the species abundance can be explained by a single component. The second axis explained a further 23% of the remaining variation in the plant species abundance.

Sample scores were plotted on the first and second axes (Figure 3). There have been some changes in the aquatic flora of Loch Dee from 1904 to 1990. The main change was between 1904 and 1983, due to the loss of *Potamogeton polygonifolious* Pouret, *Utricularia intermedia* and *U. vulgaris* L. Between 1983 and 1986 further changes in the aquatic flora were observed. However, more importantly, it appears from the PCA diagram that the aquatic flora sampled in 1990 became closer to the flora sampled by West in 1904. The aquatic plant community was more similar to the species composition of 1904 in 1990 than in the previous surveys in 1983 and 1986. This is primarily due to the recolonisation by *Utricularia* between the 1986 and 1990 surveys. The species score of *U. intermedia* pulls the 1990 sample score closer to the 1904 sample score (here assuming the species recorded as *intermedia* in 1904 and as *stygia* in 1990 are indeed synonymous).

DISCUSSION

Possible reasons for the observed within loch variation in community structure are differences in exposure to wind induced wave action, and the consequent effects of this on substrate particle size distribution (Murphy, *et al.*, 1986).

Murphy *et al.* (1986) considered that there was evidence for an increase in the population size of *Juncus bulbosus* in Loch Dee since West's survey. This species is commonly recorded as abundant in acidified water (Arts, Roelofs & De Lyon, 1990). West did not mention this plant as being worthy of note in Loch Dee in 1904-5 (although elsewhere for example Loch Enoch, it was noted to be very abundant). By 1986 *J. bulbosus* was the third most common species in Loch Dee occurring in >50% of the macrophyte samples. There was no evidence in any of the surveys of deep water colonisation by *Sphagnum* spp., which is a characteristic feature of many other acidified lakes (Grahn, 1985).

The 1990 plant survey provides initial evidence of changing macrophyte community structure in Loch Dee particularly in the submerged vegetation, with a recent return to a flora closer to that recorded by West in 1904. The return of *Utricularia,* in some abundance is of particular interest as a similar response to liming of acidified waters has been reported in other loch deacidification studies such as Loch Fleet, Scotland (Battarbee *et al.*, 1991) and Lake Trehörningen in Sweden (Eriksson, Hörnström, Mossberg & Nyberg, 1983).

CONCLUSION

The macrophyte record is clearly incomplete with no records for the intervening 79 years between 1904 and 1983. However, we suggest evidence for a reduction in species diversity between 1904/5 and the early 1980s. After the implementation of a selective liming programme in 1983 and 1985 (see Maucotel & Werritty, this volume) the species richness of submersed aquatic macrophytes in Loch Dee increased and the flora returned to a similar state as recorded in 1904/5 by West.

ACKNOWLEDGEMENTS

The authors are indebted to Ms Aileen Adams for technical assistance during the 1986 & 1990/91 field work. The 1986 survey was carried on contract to Department of Agriculture and Fisheries for Scotland, Freshwater Fisheries Laboratory, Pitlochry. The 1990/1991 work was funded by the Science & Engineering Research Council (Grant No. 89303645). Thanks are also due to Dr Andrew Spink from The University of Utrecht for reviewing the text.

REFERENCES

Ali, M M, 1992. *Ecological studies on freshwater macrophytes in regulated waterbodies in Egypt and U.K.* Unpublished PhD thesis University of Assiut, Egypt.

Arts, G H P, A J de Haan, M B Siebum, G M Verheggen, 1989. Extent and historical development of the decline of Dutch soft waters. Proceeding of the Koninklijk 3 Nederlandse Akademide van.. Ecology, Proceedings C., **92,** 281-295.

Arts, G H P, J G M Roelofs, M J H De Lyon, 1990. Differential tolerances among soft-water macrophyte species to acidification. Canadian Journal of Botany, **68,** 2127-2134.

Battarbee, R W, N A Logan, K J Murphy, P Raven, R J Aston, G N Foster, 1991. Other Aquatic Biology. In: *Restoring Acid Waters: Loch Fleet 1984-1990* (eds: G Howells and T R K Dalziel), pp289-330. Elsevier Applied Science, London.

Den Hartog, C, C Segal, 1964. A new classification of water plant communities. Acta Botanica Neerlandica, **13,** 367-393.

Ekman S, 1911. Neue apparate zur qualitativen und quantitaven erforschung der bodenfauna der seen. Int. Rev. Ges. Hydrobiol, Hydrogr., **3,** 553-561.

Eriksson F, E Hörnström, P Mossberg, P Nyberg, 1983. Ecological effects of lime treatment of acidified lakes and rivers in Sweden. Hydrobiologia, **101,** 145-164.

Farmer, A, D H N Spence, 1986. The growth strategies and distribution of isoetids in Scottish freshwater lochs. Aquatic Botany, **26,** 247-258.

Farmer, A, 1990. The effects of lake acidification on aquatic macrophytes - a review. Environmental Pollution, **65,** 219-240.

Gauch, H G, 1982. *Multivariate Analysis in Community Ecology.* Cambridge University Press, Cambridge. 298pp.

Grahn, O, 1985. Macrophyte biomass and production in Lake Gårdsjön - an acidified clearwater lake in SW Sweden. Ecological Bulletins, **37,** 203-212.

Hiley P D, J F Wright, A D Berrie, 1981. A new sampler for stream benthos, epiphytic macrofauna and aquatic macrophytes. Freshwater Biology, **11,** 79-85.

Last, F T, 1989. Acidic Deposition: Case Study Scotland. In: *Acidic Precipitation Volume 1: Case Studies.* (eds: D C Adriano, M Havas). pp237-274. Springer-Verlag.

Marrs S J, K J Murphy, P J Dominy, 1993. Relationships between submerged macrophytes and algae in freshwater lochs of differing trophic status. Journal of Aquatic Plant Management (in press).

Morris, J T, 1991. Effects of nitrogen loading on wetland ecosystems with particular reference to atmospheric deposition. Annual Review of Ecology and Systematics, **22,** 257-279.

Murphy K J, S Miller, K Anderson, 1986. *Aquatic Vegetation of Loch Dee, Galloway.* Unpublished report to DAFS Freshwater Fisheries Laboratory, Pitlochry.

Raven P J, 1985. The use of aquatic macrophytes to assess water quality changes in some Galloway lochs: an exploratory study. Working Paper no.9, Palaeoecology Research Unit, University of London.

Ter Braak, C J F, 1988. *CANOCO - a FORTRAN programme for canonical community ordination by [partial] [detrended] [canonical] correspondence analysis, principal components analysis and redundancy analysis (version 2.1)* Technical report: LWA-88-02. Groep Landbouwwiskunde Wageningen, The Netherlands.

West G, 1910. A further contribution to the comparative study of the dominant phanerogamic and cryptogamic flora of aquatic habitats in Scottish lakes. Proceedings of the Royal Society of Edinburgh series B, **30,** 65-81