# The Interaction of Environmental Variables and their Influence on Macroinvertebrate Distribution within a Shallow Calcareous Lake

**By**

**Barrie Tuite**

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**Supervisor Dr Kenneth Irvine**

**Student Number: 08260993**

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## DECLARATION

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Barrie Tuite

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## Abstract

Extensive work has been carried out on macroinvertebrates in lotic systems but far less information is available on their ecology in lakes. Benthic macroinvertebrates play a key role in the functioning of lake ecosystems. Changes in this functioning should therefore be reflected by changes in the macroinvertebrate community. In order to discriminate between natural and human-induced variation in macroinvertebrate communities, the factors which influence their distribution must be determined.

Environmental variables rarely act in isolation on biotic communities. It is often the specific combination of factors which will determine the nature of pressure applied to invertebrate assemblages. This project investigated the combined and individual effects of a range of chemical and hydromorphological variables on macroinvertebrate distribution. Evidence of interdependence among variables was examined and the possible impact of such interdependence on invertebrate distribution was considered. The work was carried out on Lough Carra in Co. Mayo which is one of the finest examples of a shallow calcareous low-nutrient lake in Europe. Extensive research over the past decade has found evidence of declining ecological conditions within the lake.

The biotic community was characterised by the dominance of the mayfly species *Caenis luctuosa* and the low abundance of pollution-sensitive species. The results of this project show that depth had a strong influence on invertebrate assemblages but only at sites deeper than 4 m. Aside from depth, no relationship was found between any variable, or combination of variables, and macroinvertebrate distribution. If any such influence does exist, research with a narrower focus may be necessary to detect it.

A strong positive correlation was found between depth and all metals and nutrients measured. This relationship should be considered when comparing chemical values within and among lakes. Total phosphorus was strongly correlated to iron and manganese, owing to binding of phosphate to metals which reduces bioavailability of phosphate. Comparison of the results with previous studies suggests an increase in total phosphorus and a reduction the nutrient binding capacity of the sediment, since 2005. The potential exists for phosphate saturation of the sediment leading to nutrient enrichment of the lake; remediation measures would be hampered by natural feedback mechanisms.

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# 1. Introduction

## 1.1 Project Rationale

In order to successfully manage and protect lake ecosystems, knowledge of factors influencing their ecology is essential. The biotic community present in a lake is determined by a wide array of environmental variables. Water depth, pH, nutrient content and hydromorphology all form part of the abiotic framework, within which, the biotic community exists. An alteration to these environmental variables can result in pressure being exerted on the biotic community. If a species within the community cannot adapt to the pressure, their ability to reproduce and compete for resources will be adversely affected. A change in environmental variables should therefore lead to some change in the biotic community structure.

Detecting the exact nature of community alteration caused by environmental factors is a difficult task. Attempts to study, in isolation, the influence of one variable can yield only limited results. Factors such as nutrient level, water depth and metal concentration do not act alone on biological communities. It is often the interaction of these variables which will determine the exact nature of the pressure imposed on communities. The situation is further complicated by the response of biotic communities to pressure. Many feedback mechanisms exist and the response can alter the nature of the pressure itself e.g. nutrient loading causes increase in phytoplankton growth, which in turn causes a drop in nutrients.

The traditional approach to assessing freshwater ecosystem quality has involved chemical analysis of the water column for compounds such as phosphorus and chlorophyll-*a*. Such testing will give a snap-shot of the current water condition but give little information on long-term changes in the lake ecosystem. If, for example, a lake is subject to heavy metal contamination, the pollutant may precipitate rapidly and so, although the water column can appear clean, high metal concentrations may be present in the lake sediment. Nutrients may also be present at high concentrations in sediment but not in the water column above. Release of these nutrients owing to changing chemical conditions could dramatically alter lake ecology.

It is clear that chemical analysis of sediment can give valuable information regarding pressures exerted on a lake but, in order to present a more complete picture of ecological status, the effects of these variables on the ecosystem must be assessed. Macroinvertebrates represent an important component of lake ecosystems. They are defined by the absence of a spinal column and above microscopic size. Benthic macroinvertebrates are those found on or in the lake bottom sediment. Their distribution and ecology has been well studied in rivers but far less work has been done on their ecology in lentic systems. Macroinvertebrates play a vital role in food webs, and knowledge of their ecology aids understanding of interactions at higher and lower levels of the food chain. Aside from their role in food webs, macroinvertebrates can be used as bioindicators of ecological status in aquatic systems. If damage to lake ecosystems is to be detected through macroinvertebrates, the factors which influence their distribution must be studied.

In this project a suite of environmental variables (total phosphorus, total nitrogen, magnesium, manganese, iron, calcium, water depth and substrate type) were analysed from sediment at a variety of sites within Lough Carra, a shallow, calcareous lake in the west of Ireland. Relationships among these variables were examined. These variables were then compared with the macroinvertebrate community sampled at each site. Evidence of relationships between the biotic and abiotic data was investigated. Studies of Lough Carra over the past decade have charted a general decline in ecological status owing to nutrient enrichment. If factors such as depth and sediment chemistry affect nutrient behaviour in the lake, these factors must be considered when analysing data from sites within the lake and when comparing Lough Carra with other lakes. Furthermore, knowledge of how these interactions affect the biotic community is necessary to detect, and mitigate against, anthropogenic alteration of the Lough Carra ecosystem.

Three different methods were used to sample macoinvertebrates within the lake. These methods were compared in terms of total abundance and species richness yielded from each.

### 1.2. Bioindicators

Bioindicators have been used in the assessment of aquatic ecosystems since the early 20$^{th}$ century (Rosenberg and Resh, 1993). To successfully use benthic macroinvertebrates as bioindicators, a specific pressure-load-dose-response must be identified. The theory being that when an environmental pressure impacts on a habitat, the ability of some macroinvertebrate species to survive and reproduce will be hindered. The area affected by the pressure will display a reduction in the abundance of these species as they either move away or die off. The abundance of species capable of withstanding the pressure will increase and so the macroinvertebrate community structure will alter.

If macroinvertebrates are to be used as bioindicators across a broad range of freshwater habitats, the factors which limit their use must be considered. The sensitivity of macroinvertebrates to organic pollutants varies greatly among and even within families. For example the Chironomidae species *Chironomus plumosus* and *Chironomus riparius* are largely insensitive to severe organic pollution, while *Brillia longifurca* of the same family is intolerant to even low levels of nutrient enrichment. The use of bioindicators is further complicated by the fact that individual families display varying tolerance to different pollutants e.g. Gastropods of the family Physae are tolerant of organic pollution but sensitive to copper. In the case of many Plecoptera species the opposite is true (Gray, 2005). This variation in sensitivity highlights the desirability of species level identification when using macroinvertebrates as bioindicators.

Previous studies have shown that a wide variety of macroinvertebrates can be found in Lough Carra, including Ephemeroptera, *Asellus* spp. and *Gammarus* spp. These taxa display a range of nutrient and substrate preferences at species level. The majority of the species within the order Ephemeroptera require stable substrate for burrowing and are sensitive to organic pollution but some of the Caenidae and Baetidae species can tolerate severe levels. *Asellus* spp. are found across a range of trophic conditions and substrate types. While many *Gammarus* spp. are associated with low nutrient concentration waters, some species (e.g. *G. pulex*) can adapt to relatively high organic pollutant levels (Moss, 2000, Džeroski *et al.,* 2000, Gray, 2005).

An important element to consider when surveying macroinvertebrate larval communities is that the absence of a pollution-sensitive species does not necessarily imply pollution. The area sampled may represent an unsuitable mesohabitat for that species owing to factors such as macrophyte cover or water depth. The environmental conditions present in Lough Carra (high alkalinity, shallow water, abundant fine-grained sediment) have been highlighted as strong influences on macroinvertebate distribution in other lakes.

### 1.3 Macroinvertebrate Sampling Methods

A wide variety of methods may be used to sample benthic macroinvertebrates in freshwater ecosystems. Net sweeping, where a net is passed through macrophyte cover a set number of times, is a commonly used method in shallow water areas. Kick sampling may also be used, in this case the sample area is agitated by the operator’s feet and a net is passed through the resulting disturbed sediment. In areas too deep for kick sampling the net may be moved over the sediment surface from a boat. Suction sampling uses a flexible tube attached to a suction pump on board a vessel. The nozzle of the tube is held in contact with the sediment surface and a sediment/water mixture is pumped into a container from which it is then sieved.

None of these methods are feasible for use in deeper water sites (2 m + approx.) where handling a net becomes impractical and tubing cannot be kept in contact with the sediment in a controlled manner. In deeper areas, a form of dredge (e.g. Eckman grab) is often used. These apparatus consist of steel box with spring-loaded metal jaws which is lowered on a line to the bottom of the waterbody. A weight is then sent down the line which releases the jaws upon impact with the grab. A sample of sediment is thereby contained in the grab, which is then hauled to the surface.

Many lake surveys require sampling at a range of depths and so a variety of sampling techniques must be employed. Knowledge of the selectivity of these different techniques greatly aids comparison among them. Previous work has shown that net sampling can retain a different sub-set of the macroinvertebrate community to grab sampling (Humphries *et al.,* 1998, McGoff and Irvine, 2009). Burrowing animals may be over represented by grab sampling, while the opposite can occur with net samples. Net and grab sampling techniques have been well-studied, at least in lentic systems, but very little work has been done comparing these methods to suction sampling. The suction method may be very useful fpr sampling sediment in boulder-strewn areas. In this study, the macroinvertebrate abundance and species richness yielded from three methods (net, grab, pump) was compared. It is hoped that the data generated will aid decision-making regarding selection of sampling method for specific sites, and allow for more accurate comparison of results among the three techniques.

## 1.4 Factors Affecting Benthic Macroinvertebrate Distribution

### 1.4.1 Water Depth

It has been well established that the littoral and profundal lake zones support different benthic macroinvertebrate communities (Moss, 2000). This difference is largely the result of lower dissolved oxygen levels at deep water sites. In deep water, the lake bottom receives less light than in shallower areas and so the rate of photosynthesis and accompanying oxygen production is lower and in the profundal zones ceases completely. Bacterial decomposition of organic matter still takes place at these deep water sites, which can deplete the already low oxygen levels.

While the lake chosen for sampling in this project is shallow, with a mean depth of approximately 1.75 m, differences in macroinvertebrate assemblages have been noted within the littoral zone of many lakes. Furthermore, the maximum depth of Lough Carra is over 18 m and so the potential does exist for a broad range of depths to be sampled. Several studies have reported a reduction in taxa richness with increasing water depth within the lake littoral zone (Czachorowski, 1993, Graca *et al.,* 2004, Baumgartner *et al.,* 2008). Rieradevall *et al.* (1999) also reported a fall in taxa richness with depth in the littoral zone and attributed this to a reduction in dissolved oxygen, lower food quantity/quality and a change in substrate.

Nutrient enrichment of lakes can cause low oxygen conditions, which is also associated with deeper water. Establishing what biotic communities are associated with a particular depth range can help determine if the invertebrate assemblage recorded at a site is owing to nutrient enrichment or simply the natural influence of water depth.

### 1.4.2 Substrate Type

A broad range of research has shown that sediment characteristics have a significant impact on benthic macroinvertebrate taxa richness and diversity. Several studies have found specific taxa associated with specific sediment (Dermott, 1978, Rieradevall *et al.,* 1999, Stoffels *et al.,* 2005, Brauns *et al.,* 2007). Some authors have concluded that larger particle size gives greater taxa richness (Heino, 2000, Graca *et al.,* 2004). While larger particles may support greater taxa richness, small grained sediment can contain higher taxa abundance. A study by Casorbi *et al.* (2002) found that fine-grained silt supported higher taxa abundance than sand or gravel. This may be owing to reduced competition, allowing some species of Chironomidae and Oligochaetae to reproduce in high numbers, while burrowing species such as Ephemeroptera are excluded owing to the lack of sediment structural integrity. Studies in upland streams support this theory, where an increase in fine sediment was shown to reduce taxa richness and diversity of Ephemeroptera, Plecoptera and Trichoptera (Kaller *et al.,* 2001).

Sediment comprising varied particle sizes has been found to support greater taxa richness but low total macroinvertebrate biomass (Wohl *et al.,* 1995). The findings of Gayraud *et al.* (2001) conflicted with those of Wohl *et al.* (1995) with no greater taxa richness found in complex substrate but this may have been owing to the narrow range of substrate types sampled.

The absence or presence of macrophytes has been shown to strongly influence macroinvertebrate assemblages with most studies finding higher taxa richness and abundance in areas with macrophyte growth. This was also the case with work by White and Irvine (2003) and Della Bella (2004) but they also found that fine sediment areas free of macrophytes contained high densities of macroinvertebrate species which were not found in other mesohabitats of the same lake. Their conclusion was that sites free of macrophytes were more suitable for monitoring bioindicators as they are easier to sample and sort and are likely to contain distinct species.

Research results have varied regarding the influence of habitat on taxa richness and abundance. There is a perception among some researchers that lake mesohabitats are so variable that meaningful comparisons of benthos among different lakes cannot be made. A study by in 2001 on three basins within one lake in Finland concluded that habitat structure had a stronger influence on macroinvertebrate assemblages than other environmental factors (Tolonen *et al.,* 2001). The opposite was found by several other studies (White and Irvine, 2003, Trigal *et al.,* 2006, Trigal *et al.,* 2007).

These conflicting results suggest that the complex interaction of physical and chemical parameters can result in different substrate and fauna interactions in different lakes. Area specific research may be necessary to determine what effect substrate has on invertebrate distribution within a specific lake, as general guidelines for other waterbodies may not apply.

### 1.4.3 Organic Matter Content

While the majority of organic matter in a lake may be present in the form of living macrophytes, very little of this material is utilised directly by macroinvertebrates i.e. few species graze directly on vegetation (James *et al.,* 2000, Moss, 2000). Macroinvertebrates may feed as grazers on the periphyton which grow on the macrophytes (e.g. Mollusca), as shredders or collectors of organic particles (e.g. some Trichoptera) or as predators feeding on other invertebrates (e.g. Odonata).

The level of organic material in aquatic substrate can therefore have a significant influence on the macroinvertebrate assemblages present. Some recent studies in both lentic and lotic systems found organic matter content to have the strongest the influence on macroinvertebrate distribution when compared with variables such as substrate, vegetation cover and pH (Yo Miyake, 2002, Syrovatka *et al.,* 2009, Takamura *et al.,* 2009). The study by Takamura (2009), in a shallow lake found high abundance of *Tubificide* species and *Tanypus* species of Chironomidae in substrate with high organic matter content (OMC) while *Sphaerium spp*. and the Chironomidae species *Monodiamesa* were associated with low OMC substrate. Work by Miyake *et al.* (2002) found higher total macroinvertebrate taxa richness and abundance in high particulate organic matter substrate. A study in 2004 by Fuller *et al.,* found that increasing organic carbon altered community composition in freshwater systems and that the form this carbon took, determined the effect it would have, owing to selective feeding by some invertebrates. A higher concentration of bacteria lead to increased numbers of Chironomidae, while increased levels of algae produced a higher density of Ephemeroptera.

In lotic systems terrestrial input of organic matter is a major source of energy and reducing this input can significantly reduce macroinvertebrate species abundance and richness (Wallace *et al.,* 1999). In contrast, large lentic systems receive the majority of carbon from autochthonous sources and reduction in allochtonous biomass appears to have little effect on food webs (Rubbo *et al.,* 2008). High primary production within lakes is the most likely source of high organic carbon in the substrate. However, high primary productivity is dependent on nutrients, (mainly phosphorus) which enters the lake from the catchment.

### 1.5 Lake Eutrophication and Macroinvertebrates

In recent decades, eutrophication has been recognised as a major threat to freshwater systems in Ireland. Land use change and more intensive agriculture have lead to higher nutrient levels entering ground and surface waters (Smith *et al.,* 2005). Higher nutrient levels result in increased primary production within water bodies, which is often noticeable within the water column as algae blooms. Macrophyte biomass may also increase initially but when vegetation dies it sinks to the bottom of the water body and is aerobically decomposed. This reduces the dissolved oxygen concentration of the water. If nutrient input to a water body is sufficiently prolonged or concentrated, the oxygen level will become too low for many fish, macroinvertebrate and macrophyte species to tolerate, resulting in an overall reduction in species diversity (Moss, 2000).

Nitrogen and phosphorus are the nutrients which play the most important role in eutrophication, as primary producers have a high demand for these, relative to the amounts available in the environment. Phosphorus enters water bodies in run-off from the catchment via atmospheric deposition and may also be released from the lake sediment. Nitrogen also enters water bodies in run-off, precipitation and through fixation of atmospheric nitrogen by some cyanobacteria and bacteria. In freshwater systems, phosphorus is generally the limiting nutrient for primary producers i.e. the level of nitrogen present is in excess of the organisms' requirements, while it is the amount of phosphorus present which is preventing further growth. In such situations, an increase in phosphorus will lead to an increase in biomass (Schindler 1974).

Lakes are particularly sensitive to increased nutrient levels owing to the limited mixing and long retention time of the water body. Many macroinvertebrate species are known to be sensitive to organic enrichment and are widely used in assessment of running water quality. However, efforts to qualify or quantify the relationship between macroinvertebrate assemblage and lake trophic state have produced conflicting results.

Some macroinvertebrate species have been linked to specific lake trophic status e.g. most Ephemeroptera, plecoptera, trichoptera are associated with oliogotrophic or mesotrophic lakes. However great variation in sensitivity has been found at family and even genus level e.g. within the chironomids. Abundance of individual species may not be indicative of total biodiversity and sampling a very broad range of species is impractical. The challenge is to find widely distributed species which are more strongly influenced by trophic status than other environmental variables.

The study by Tolonen *et al.* (2001) found that substrate type had a greater influence on littoral macroinvertebrate community structure than trophic status but a narrow range of trophic states where sampled in the study. More recent work by Brauns *et al.* (2007), on a wider range of trophic states also concluded that trophic status influences macroinvertebrate community structure but not as strongly as habitat type. Several authors have highlighted the difficulty in comparing research on different lakes as many studies refer to lakes as oligotrophic, mesotrophic and eutrophic but do not give specific concentrations of total phosphorus or chlorophyll-*a* (O'Toole *et al.,* 2007).

The relationship between macroinvertebrates and eutrophication is complex and at present, poorly understood. Analysis of a large data set complied from across the European Union concluded that the macroinvertebrates species examined in that study, could not, at present be used as practical indicators of lake trophic status (O'Toole *et al.,* 2007). Further study is required on the interactions among macroinvertebrates, trophic status and other environmental variables such as macrophyte cover, periphyton biomass and substrate type.

### 1.6 Sediment and Nutrient Interactions

The discussion above highlights the effects of increased nutrients on lake ecosystems. However, when attempting to predict these effects, it is not just the total quantity of nutrients within the water body must also be taken into account, but also form these nutrients take . Phosphorus is usually the limiting nutrient in freshwater systems, where its most bioavailable form is dissolved inorganic soluble reactive phosphorus. Phosphorus can be readily utilised by primary producers only when it is freely dissolved in the water as phosphate ions. Phosphate ions have a high affinity for iron and manganese ions (Löfgren and Boström, 1989, Gunnars *et al.,* 2002, Griffioen, 2006). Phosphate bound to these metals is not immediately bioavailable and so will not provide nutrition for primary producers (Ellison and Brett, 2006).

Phosphate/metal interactions are complex and dynamic with bonds forming and breaking as environmental conditions change. pH plays a major role in the formation of phosphate/iron complexes. At low pH, phosphate is tightly bound to metal ions but at high pH (approx. pH 8), hydroxide ions are exchanged with phosphate ions at the binding sites on metals (Jin *et al.,* 2006). This releases the phosphate ions into the water column, making them bioavailable.

One of the most important factors governing phosphate/iron binding is oxygen availability. In well-oxygenated water, dissolved iron is oxidised to form insoluble $Fe^{3+}$ ions which bind with phosphate and precipitate through the water column to sediment surface; the bioavailability of this phosphate is significantly reduced. If oxic conditions persist on the sediment surface, the phosphate will remain bound to the metal. However, organic matter from the water column settles on the sediment surface and is decomposed by bacteria, utilising oxygen. In areas where photosynthesis levels are low (e.g. deep water sites), little oxygen is produced to replace that utilised by the bacteria and the sediment/water interface can become anoxic. Under these anoxic conditions, iron is reduced, forming $Fe^{2+}$, and the phosphorus is released back into the water column as bioavailable phosphate (Bronmark and Hansson, 2005, Ellison and Brett, 2006).

Recent work on Lough Carra by Hobbs *et al.,* (2005) showed an increase in total phosphate in the upper 10 cm of the lake sediment and a decrease in the ratio of iron to phosphate in the same sediment section. This suggests that the sediment is becoming saturated with phosphate, preventing further binding. If phosphate input continues at its current rate, the buffering capacity of the lake sediment may be lost, and far more of the phosphate entering the lake will remain bioavailable. This would lead to increased primary productivity and subsequent eutrophication. Eutrophication can often result in anoxic conditions which would increase the amount of phosphate released from the sediment and exacerbate the eutrophication. A natural feedback mechanism such as this would be very difficult to reverse (Irvine *et al.,* 2003, Hobbs *et al.,* 2005).

### 1.7 Lake Description

Lough Carra is located in Co. Mayo in the west of Ireland. The lake lies within the Lough Corrib catchment which has an area of approximately 104 $km^2$. It is a shallow (mean depth 1.75 m), hardwater (>100mg $1^{-1}$ $CaCO_3$), lowland lake (21 m asl) with a surface area of approximately 14.4 $km^2$ (King and Champ, 2000). The bedrock of the lake and its catchment is predominately carboniferous limestone overlain with limestone glacial till soils. Calcium carbonate precipitate from the bedrock is deposited on the lake bed in the form of fine-grained marl.

The lake is an important conservation site for a number of reasons. As a lowland, calcareous lake it is classified as a Special Area of Conservation (SAC 001774) under the EU Habitats Directive. The lake catchment contains protected habitats such as limestone pavement and *Cladium* fen and a wide variety of rare orchid species. The lake is also listed as a Special Protected Area (SPA 004051) under the EU Birds Directive and supports wintering populations of shoveller and gadwell (www.npws.ie). Lough Carra has been managed as a brown trout fishery since 1956 and is one of last wild brown trout calcareous lakes in Europe (King and Champ, 2000, Irvine *et al.,* 2003).

Despite the lake's protected status and considerable ecological value, its quality has deteriorated in recent years owing to anthropogenic pressure. Water quality analysis in 1997 classified the lake as oligotrophic (McCarthy, 1997). The lake is defined as mesotrophic by the National Parks and Wildlife Service (www.npws.ie) but the Environmental Protection Agency has upgraded its status to oliogotrophic in their most recent report (www.epa.ie). Despite the EPA report, recent studies show a gradual decline in water quality and a build-up of phosphorus (P), with work by Irvine *et al.* (2003) recording mean total P at 10.4 $\mu g\ l^{-1}$ ($\pm 1.3\ \mu g\ l^{-1}$).

Analysis of sediment cores taken from the lake in 2002 showed an increase in total phosphorus loading in recent decades (Hobbs *et al.,*). The exact effect of this increased nutrient input on the biota of the lake is, as yet, unknown. No records exist of the lake's invertebrate community prior in nutrient enrichment so comparison of that community with the current biota is not possible. The current macroinvertebrate community can be compared with those of reference lakes which are considered to be minimally impacted by human activity. Such comparisons are obviously hampered by natural variation among lakes. Several macroinvertebrate surveys have been carried out on the lake in recent years but for many of these only a limited number of habitats were sampled. Some surveys did record invertebrate communities dominated by pollution-tolerant species with a low number of nutrient-sensitive species. Further study is required to determine if these communities are a product of human impact or natural habitat conditions.

The decline in water quality and increase in phosphorus in the lake is most likely owing to changes in land use within the lake catchment. A survey of farmers in the area showed significant land use changes in the catchment since 1970 (Huxley and Thorton, 2004). Levels of fertilizer application has increased, as has density of livestock (sheep, cattle, pigs) and an estimated 25% of landholdings in the catchment were converted from natural or semi-natural habitat to more intensive agricultural land since 1970.

### 1.8 Project Aims and Questions

Aquatic ecosystems are, at their most fundamental level, a combination of interactions. These interactions take the form of pressures and responses. Human activity is often the basis of pressure to an ecosystem and the response can be a series of complex interactions. This response may not be immediately obvious but can have far-reaching consequences. Many Irish lakes have been degraded as a result of anthropogenic activity in recent decades. Detecting and reversing this degradation can be a difficult task owing to the natural feedback mechanisms which exist in lake ecosystems. Macroinvertebrates, as key components of lakes food webs, can play an important role in highlighting alteration of such habitats.

In order to differentiate between human-induced and natural variation within macroinvertebrate communities, an understanding of the environmental variables which influence their distribution is required.

The aim of this project was to study the individual and combined influence of environmental variables on macroinvertebrate distribution within Lough Carra. Evidence of change in macroinvertebrate community structure with altering environmental conditions was investigated. As a rare and valuable habitat which is under increasing anthropogenic pressure, a deeper understanding of the Lough Carra ecosystem is essential to its' successful management. The following questions were examined:

- Is there a difference in the abundance or taxa richness yielded by different sampling methods?
- Is there evidence of depth or substrate type influencing macroinvertebrate community structure and is this influence stronger than that of nutrient concentration?
- Is there a relationship between the sediment chemistry and the concentration of total phosphorus present and is this relationship influenced by water depth or sediment type?
- Is the level of nutrients in the sediment influencing the distribution of benthic macroinvertebrates?
- Is there evidence of a change in sediment chemistry since the 2002 study of Lough Carra and is any such change reflected in the macroinvertebrate community present?

## 2. Materials and Methods

### 2.1 Field Sampling

Field sampling was performed on the 19$^{th}$ and 20$^{th}$ of April 2009. Twenty sites were sampled throughout the lake. The samples taken for chemical analysis at sites 8 and 17 were misplaced. Owing to time constraints, not all samples collected could be processed and it was necessary to prioritise samples in terms of value to the total data set. As samples 8 and 17 could not be compared with other sites in terms of chemistry or sediment type, it was decided not to process these samples.

The original sampling design involved an equal number of sites in the mid and south basin, to allow a balanced comparison of nutrient levels in the two basins. Owing to technical difficulties outlined below, only 5 sites were sampled in the mid basin, while 13 sites were sampled in the south basin.

The location co-ordinates of each sample site were recorded using a Global Positioning System. These locations were then marked on a map of the lake which allowed for examination of spatial patterns in biotic and abiotic data. The depth was recorded using a Scubapro ® PPS-2 electronic depth finder. A combination of deep (>4 m), medium (2-4 m) and shallow water (< 2 m) sites were sampled. This range of depths was chosen in an effort to detect variation in sediment chemistry or biotic communities induced by depth. Shallow sites (<2 m) generally receive abundant light, have high productivity, complex habitat and high taxa richness. The deep sites may also support photosynthesis but at a lower rate. Less water mixing than in shallow areas should result in more homogenous sediment. The mid water sites may represent the intermediate phase between the other two habitats. All sites chosen were free of macrophyte cover in order to reduce the influence of this variable on sample results. A comparison of sites with and without macrophyte cover was not feasible owing to the limited time available for sample sorting and identification.

A total of three sampling methods were used:

1) Net: 30 second sweep using zig-zag motion across the sediment surface of the lakebed using a standard Freshwater Biological Association 1mm diameter mesh net.

2) Eckman grab: A grab with a sample surface area of 225 $cm^2$ was used to take two samples from the sediment at each site. A sediment sample was scraped from the surface of the first grab sample and placed in a plastic universal tube for chemical analysis and sediment classification. The contents of the two grabs were then combined.

3) Pump: A Whale Gusher® 30 hand pump fitted with stiff 38 mm diameter plastic tubing was used to sample the lakebed (after Tolonen *et al.,*). The tubing inlet was placed on the lakebed and pumping was performed until three buckets with a combined volume of 27 litres were filled with the water/sediment mixture.

The net and pump could not reach the lake bottom at depths greater than 2 m and so were used only at shallow water sites. The Eckman grab could be used down to the maximum depth found within the lake, while the Eckman grab alone would be used at sites with water depth greater than two metres. However, on the second day of sampling, the Eckman grab broke and no further deep water sites could be sampled. A total of 14 sites were sampled prior to the grab breakage. The remaining 4 sites were sampled with the net and pump only (Table 1 shows breakdown of sampling regime, Figure 1 gives site locations).

All net, grab and pump samples were sieved through a 1mm stainless steel sieve and the retained contents were stored in a minimum 70% alcohol solution.

Table 1. Overview of sampling regime

| Site Number | GPS Co-ordinates | Mean Depth (m) | Samples Taken | Basin Sampled |
|---|---|---|---|---|
| 1 | 53.42.522 09.13.538 | 1.3 | Net, Grab, Pump | South |
| 2 | 53.42.288 09.13.343 | 1.6 | Net, Grab, Pump | South |
| 3 | 53.42.153 09.13.332 | 1.7 | Net, Grab, Pump | South |
| 4 | 53.42.270 09.13.764 | 1.4 | Net, Grab, Pump | South |
| 5 | 53.42.050 09.13.493 | 2.1 | Grab | South |
| 6 | 53.41.989 09.14.043 | 0.8 | Net, Grab, Pump | South |
| 7 | 53.41.849 09.13.380 | 4.1 | Grab | South |
| 9 | 53.41.609 09.14.155 | 1.5 | Net, Grab, Pump | South |
| 10 | 53.41.563 09.14.074 | 2.4 | Grab | South |
| 11 | 53.41.611 09.13.450 | 2.7 | Grab | South |
| 12 | 53.41.787 09.13.327 | 5.3 | Grab | South |
| 13 | 53.41.744 09.13.649 | 2.6 | Grab | South |
| 14 | 53.41.503 09.13.994 | 2.8 | Grab | South |
| 15 | 53.42.352 09.15.720 | 1.3 | Net, Grab, Pump | Mid |
| 16 | 53.42.759 09.15.734 | 1.5 | Net, Pump | Mid |
| 18 | 53.43.106 09.16.603 | 1.5 | Net, Pump | Mid |
| 19 | 53.42.951 09.16.507 | 1.5 | Net, Pump | Mid |
| 20 | 53.42.726 09.16.387 | 1.6 | Net, Pump | Mid |


Figure 1. Map of Lough Carra, Co. Mayo showing sample site locations

## 2.2 Macroinvertebrate Sorting and Identification

Net, grab and pump samples were sorted to remove all macroinvertebrates present. Samples were first drained of alcohol using a 500 µm sieve. The sieved contents were placed in a sorting tray with water and inspected visually. If the total macroinvertebrate abundance was estimated to be below 200 specimens, the entire sample was sorted. If the total abundance was estimated to be above 200, sub-sampling was performed. To do this,the sediment sample was drained and weighed in a sieve of known weight. The sediment was then placed in a 1 litre beaker with approximately 1 litre of water. The sample was stirred to suspend the sediment and then poured rapidly into the funnel of the sub-sampler. The sediment/water mixture left the funnel through two spouts and was collected under each spout in a separate beaker. One of these beakers was selected randomly and the contents were drained and weighed. The weight of this sediment was calculated as a percentage of the total sample. If this sub-sample comprised between 40% and 60% of the total sample weight, it was considered representative of the total sample and used for sorting and identification. If the sub-sample weight was not in the 40% to 60% range, the two sub-samples were combined and the procedure was repeated until the desired weight range was achieved.  The sub-sample was then processed in the same manner as all other samples. All macroinvertebrates removed from each sediment sample were placed in glass vials with 80% industrial methylated spirits. The specimens from each sorted sample were counted and identified using the following keys: Macan, 1977, Elliot and Mann, 1979, Elliot *et al.,* 1988, Friday, 1988, Wallace *et al.,* 1990, Gledhill *et al.,* 1993, Fitter and Manuel, 1994. An Olympus SZ30 stereomicroscope was used for examination of specimens.

Oligochaetae were identified to order. Coleoptera larvae, Chironomidae, Ceratopogonidae and Sphaeriidae were identified to family level. All other specimens were identified to species level where possible. For sub-samples, the count for each species was extrapolated, using the weigh of the sub-sample as a percentage of total weight, to estimate the count for the full sample.

## 2.3 Chemical Analysis of Sediment

The sediment samples taken for chemical analysis were frozen on the 21st April 2009. Chemical analysis took place between 15th and 21st June 2009.

### Classification of Sediment Type

Each sediment sample was spread in an aluminium tray and inspected visually. The size and relative portion of the grains present was estimated. Each sample was then classified according to increasing dominance of large particles. Eight different sediment classes were identified and each allocated a number (Table 2).

Table 2. Classification of sediment based on increasing dominance of large particles

| Dominant Particle | Code |
|---|---|
| All marl | 1 |
| All silt | 2 |
| All sand | 3 |
| Marl / Gravel Mix | 4 |
| All gravel | 5 |
| Fine aggregated marl | 6 |
| Agg. Marl / Pebble | 7 |
| All course agg. Marl | 8 |

### Chemical Analysis

Samples were oven-dried at $60^0C$ overnight, then ground using a pestle and mortar. These ground samples were sieved through a 2 mm stainless steel sieve and dried at $100^0C$ overnight. The dried samples were then ground to a fine powder using a mechanical grinder. After grinding, the samples were again dried at $100^0C$ for 1 hour and then allowed to cool in a desiccator.

The finely ground samples were tested for total phosphorus (TP), iron (Fe), magnesium (Mg), manganese (Mn) and calcium (Ca) using microwave nitric acid digestion and Coupled Plasma- Optical Emission Spectroscopy (ICP-OES). Approximately 1 gram of each sediment sample was weighed onto singleply tissue paper using a Mettler Toledo PB303 Delta Range, and the exact weight recorded. The sample was placed in a microwave sample tube with 2 ml of de-ionised water and 10 ml of 57% HCl. This solution was left in a fumehood overnight and 38 ml of deionised water was then added. The samples were digested in a Milestone Ethos Ez Microwave for two hours. The digested material was allowed to cool and then filtered through Whitman filter paper. A 20:1 dilution of the filtered sample was prepared with deionised water, using a Microlab® 500 automatic diluter. Diluted samples were then loaded on the ICP-OES. Blanks containing water and acid only and tissue, water and acid, were run. Five standards were also run on the instrument.

The 1 in 20 dilution was necessary to bring the high calcium content within the range of the standards, but the values obtained for Mg, Mn, Fe and TP were out of the range of the standards with this dilution. A second set of the same samples were ran without any dilution and tested for Mg, Mn, Fe and TP.

Analysis of total nitrogen and total organic carbon was carried out for each sample, using an Elementar elemental analyser. Approximately 60 mg of finely ground, dried sediment was weighed into a silver cup using a Mettler Toledo MX5 balance and the exact weight recorded. 100 µl of de-ionised water was added and samples were placed in a dessicactor, with a glass beaker containing approximately 50 ml of 57% HCl, for 4 hours. The samples were then removed from the dessicator and oven dried at $60^0$C, overnight. The dried samples were then tightly compacted using a sample press and loaded on the Elementar analyser. Standards and two blanks, consisting of empty silver cups were also run.

## 2.4 Data Analysis

### 2.4.1 Calculation of Molar Ratios

The molar ratio of iron to total phosphorus in sediment was calculated to give an indication of the sediment phosphorus absorption capacity. Iron plays an important role in binding phosphorus to sediment. A low ratio (close to one) indicates that few free iron molecules remain in the sediment i.e. the sediment is nearing phosphorus saturation.

### 2.4.2 Data Transformations

The total abundance and species richness values for grab samples and for combined net and pump samples were normally distributed and so required no transformation, nor did the total abundance values for *C. luctuosa*. The *Asellus aquaticus* abundance values for combined net and pump samples were log (x) transformed to achieve normal distribution for correlation analysis.

The data for calcium, manganese, sediment type, depth and total organic carbon were all normally distributed. Total nitrogen, total phosphorus, magnesium and iron were not normally distributed and log (x) transformations were performed to achieve normal distribution for correlation analysis. For multivariate analysis these same variables were log (x+1) transformed to achieve normality. As the abiotic variables were measured in different units (e.g. mg $g^{-1}$, %, metres,), the values were normalised to give comparable, dimensionless scales.

For multivariate analysis, all biotic data was fourth root transformed to balance the contribution of common and rare species.

### 2.4.3 Univariate and Bivariate Statistical Analysis

The statistical software package Datadesk® was used for t-tests, ANOVA and correlation analysis.

ANOVA was used to compare the species richness yielded by each sampling method, at the seven sites where all three methods were used. The abundance and species richness yielded from net and pump samples was compared using paired t-tests. As no significant difference was found in taxa richness or abundance between net and pump samples, the results for these two sample types at each site were combined for the remaining analysis to give a larger sample size which would be more representative of the sample area.

Pearson's Product Moment correlations were used to test for significant relationships between specimen abundance and the following variables: depth, sediment type, logTP, logTN, logMg, logFe, Mn, Ca and TOC. The same analysis was performed for species richness with the same variables.

Samples grouped according to the three depth ranges were compared in terms of species richness and total abundance using one way ANOVA.

Sediment type was classified into three groups. Types 1,2,3 (fine sediment) were placed in group A, types 4,5,6 (mixed sediment), in group B and types 7 and 8 (course sediment) in group C. One way ANOVA was used to determine if a significant difference in abundance or richness existed among these groups. ANOVA was also used to compare these sediment groupings with *C.luctuosa* and *A.aquaticus* abundance, for grab samples.

Two-way ANOVA was used to investigate the combined effect of depth and sediment type on macroinvertebrate distribution. Analysis was performed using total abundance, species richness, *C.luctuosa* and *A.aquaticus* abundance data from the fifteen grab samples. The same analysis was not practical for combined net and pump samples, as too few samples were taken within each sediment group.

Analysis of relationships between abiotic variables was carried out using Pearson's Product Moment correlation coefficients.

Mean values and 95% confidence intervals were calculated for the chemical data to allow comparison with the work of Hobbs *et al.,* (2005)

### 2.4.3 Multivariate Analysis

**Biotic Data**

The software package Primer 6.0 was used for multivariate statistical analysis of community structure and environmental variables. As with univariate analysis, data from pump and net samples were combined and grab samples were analysed separately. Sites were first compared solely on biotic data. A rank similarity matrix of the fourth root transformed data was composed using the Bray-Curtis coefficient, where all sites are compared with each other in terms of the number of species, and the number of individuals of each species, present. A value of 100 is given for two identical samples and 0 for two samples with no species in common. Hierarchical clustering was performed using this similarity matrix to group similar sites together. These sites were labelled with the water depth to highlight if biotic samples of a similar depth were clustering together.

Non-metric multi-dimensional scaling was also carried using the Bray-Curtis similarity matrix. In this analysis the ranked similarities from the matrix are represented in a 2 or 3 dimensional ordination map. The stress value of an MDS is a measure of how accurately the values from the similarity matrix can be represented on a 2 dimensional ordination. Stress value below 0.1 is considered good and below 0.05 ideal, whereby the information from the similarity matrix can be accurately represented in a 2 dimensional plot, with very little distortion.

## Abiotic Data

Abiotic data was initially analysed independently of species data using Principal Components Analysis (PCA). This method was chosen as it is well-suited to environmental data sets with few zero values. The output is a 2 or 3 dimensional ordination with sample similarities based on Euclidian distance. Depth, sediment type and all chemical variables were used in this analysis.

## Analysis of Combined Biotic Data and Abiotic Data

The MDS ordinations generated for the biotic data were overlain with bubble plots of the abiotic variables, where the size of the bubble relates to the value of the variable. This allows a visual comparison of the association between community composition and a specific environmental variable. The biotic grab data showed some indication of a relationship with depth but it was clear that other factors were also strongly influencing the community composition (Figure 6). Visual examination of all other variables overlain on biotic data, showed no sign of relationships (see Figures 7, 10 and 11 for examples)

The BEST procedure from Primer 6.0 was used to investigate relationships between the invertebrate community structure and abiotic environmental variables. This program searches for correlations between the environmental variable and the biotic data for each site. The combinations of environmental variables which have the highest rank correlation to the biotic data are then outputted by the BEST program. The degree to which a set of environmental variables “explains” the macroinvertebrate community composition is given as a Rho value, with a range of -1 to +1. A Rho value of >0.95 is deemed close to a perfect match.

The Rho value generated by the BEST procedure cannot be used in formal hypothesis testing as the ranked correlations are not independent variables; the values generated are based on interdependent similarity calculations (Clarke and Warwick, 2001). A Global BEST test was used to test the statistical significance for the Rho value generated by BEST. Global BEST calculates the probability of generating a Rho value by random permutations which is higher than the calculated test value. The closer the Rho is to 1, the higher the probability of a statistically significant difference.

## 3. Results

### 3.1 Biotic data overview

A total of 45 species, across 23 families, were found within the lake. The highest abundance recorded for one site was 2094 specimens and the highest richness recorded was 21 species. The highest abundance for a single sample was 1496 specimens, while the greatest richness recovered from one sample was 14 species.

The most prominent feature of the biotic data is the dominance of *Caenis luctuosa* at the majority of sites sampled. At fifteen of the eighteen sites, *C. luctuosa* was the most abundant species present and at four sites this species accounted for over 70% of the total abundance (Table 2). Of the three sites with no *C. luctuosa* present, two were deep water areas and the other a shallow water site with low richness and abundance. The four most abundant taxa were *C. luctuosa, A.aquaticus*, chironomidae and gammaridae (Table 3). *Asellus aquaticus* and Chironomidae showed wide distribution, while Gammaridae were restricted to fewer sites. The invasive Gammaridae *Crangonyx pseudogracilis* was recorded at several sites. Trichoptera numbers were generally low at most sites but a wide variety of genera and species were found. Gastropod counts were low, with the exception of site 15, where 20 specimens were found in one sample. Common leech species were found throughout the lake.

Table 3.The four most abundant taxa as a percentage of the total abundance at each site, sampled in Lough Carra.

| Site | Depth | Sediment Type | *C. luctuosa* % | Chironomidae spp. % | *A. aquaticus* % | Gammaridae spp. % | Total Abundance (All samples combined) |
|---|---|---|---|---|---|---|---|
| 1 | 1.3 | 4 | 48 | 26 | 10 | 4 | 1727 |
| 2 | 1.6 | 7 | 64 | 6 | 21 | 2 | 1018 |
| 3 | 1.6 | 5 | 66 | 7 | 17 | 1 | 1901 |
| 4 | 1.4 | 8 | 73 | 10 | 5 | 7 | 1329 |
| 5 | 2.1 | 8 | 37 | 26 | 12 | 0 | 289 |
| 6 | 0.8 | 7 | 75 | 14 | 2 | 2 | 2094 |
| 7 | 4.1 | 2 | 0 | 87 | 2 | 0 | 54 |
| 9 | 1.4 | 5 | 60 | 19 | 2 | 6 | 686 |
| 10 | 2.4 | 8 | 0 | 32 | 49 | 0 | 57 |
| 11 | 2.7 | 3 | 8 | 32 | 20 | 0 | 95 |
| 12 | 5.3 | 2 | 2 | 69 | 2 | 0 | 182 |
| 13 | 2.6 | 6 | 37 | 18 | 34 | 0 | 180 |
| 14 | 2.8 | 1 | 41 | 39 | 16 | 0 | 241 |
| 15 | 1.4 | 1 | 74 | 18 | 0 | 1 | 1009 |
| 16 | 1.5 | 6 | 57 | 32 | 0 | 1 | 736 |
| 18 | 1.4 | 6 | 4 | 41 | 0 | 25 | 102 |
| 19 | 1.5 | 6 | 79 | 13 | 0 | 0 | 1421 |
| 20 | 1.6 | 6 | 8 | 55 | 5 | 3 | 284 |

### 3.2 Comparison of Sampling Methods

Total abundance was far lower with grab sampling than with the net or pump samples (Table 3). There was no significant difference in the species richness yielded by each sampling method (ANOVA, $F_{2, 18} = 2.772$ $p \leq 0.0892$).

Pump sampling gave higher mean and total abundance compared with net samples (Table 3) but the difference between the two methods was not significant (paired t-test $p > 0.05$, df =10) for richness or abundance.

Table 4. Comparison of abundance and richness from grab, net and pump samples

| Sampling method | Total abundance | Mean abundance | Mean species richness |
|---|---|---|---|
| Net (n=11) | 5189 | 472 | 10.6 |
| Pump (n=11) | 6137 | 558 | 9.5 |
| Grab (n=15) | 2082 | 149 | 7.6 |

### 3.3 Factors Influencing Macroinvertebrate Distribution

#### 3.3.1 Effect of Depth

Low abundance and particularly low species richness was recorded at the deep sites (>4 m), while great variation in abundance and richness existed among the shallower sites. The deep sites were dominated by low oxygen tolerant chironomidae and gastropods, with very few Ephemeroptera (Appendix 1). No significant correlation was found between taxa richness and depth for grab samples (Figure 2), nor between total abundance and depth for the same samples (Table 9).

No significant correlation was found between abundance or taxa richness from combined net and pump samples and site depth (Table 10).

For grab samples, no significant correlation was found between *A. aquaticus* and depth but a strong negative correlation was found between the *C. luctuosa* and depth (Figure 3). At the deep water sites (sites 7, 12), almost no *C. luctuosa* were found (Appendix 1). No correlation was found between depth and *A. aquaticus* or *C. luctuosa* for the combined net and pump sites, all of which were taken at depths below 2 m. Correlation analysis involving Gammarus was not practical as the number of sites containing this taxa was too low.

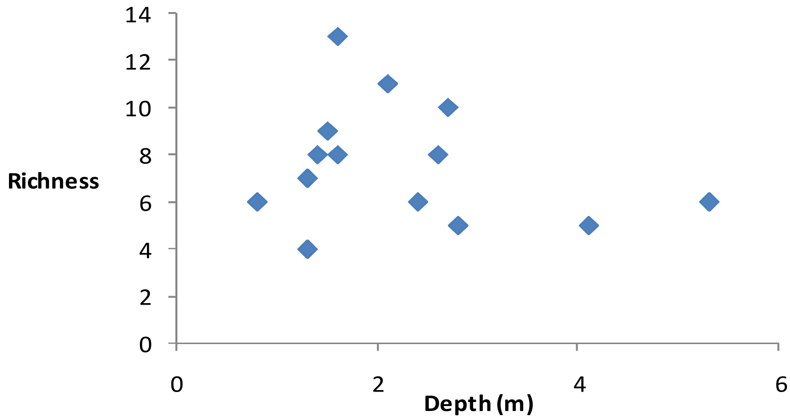


Figure 2. Scatterplot of species richness against site water depth, for grab sample data (Pearson's Product Moment Correlation for richness and depth: -0.238)

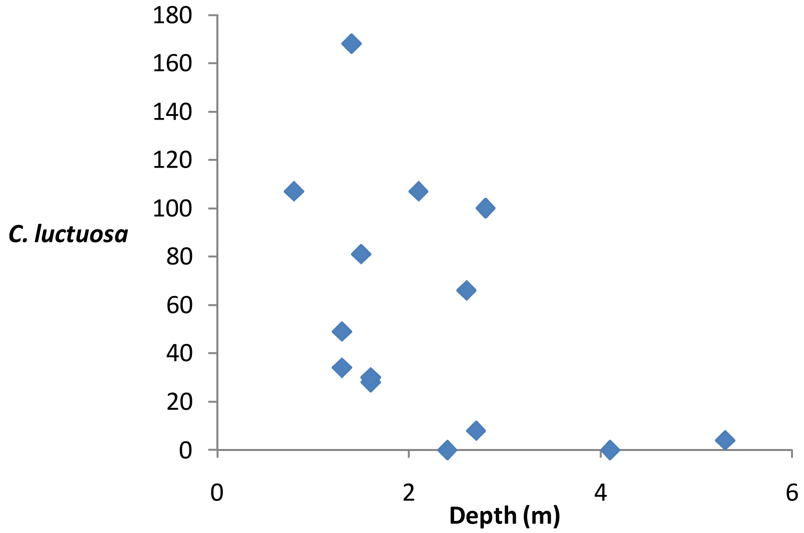


Figure 3. Scatterplot of *C luctuosa* against site water depth, for grab sample data.(Pearson's Product Moment Correlation for *C. luctuosa* and depth: -0.717)

Comparison of the three depth ranges showed no significant difference among the three groups (ANOVA $F_{2, 11} = 0.7551$, $p \leq 0.4928$ and $F_{2, 11} = 0.4083$, $p \leq 0.6744$) for richness and abundance respectively.

### 3.3.2 Effect of Substrate Type

Great variation in sediment was found across the lake, from sites with only fine-grained marl to others dominated by pebble and large pieces of solid aggregated marl. At many sites the substrate was not well-mixed and contained an approximately even mix of course and fine particles.

Table 5. Number of sites within each sediment class

| Dominant Particle | Code | No. of Sites in Category |
|---|---|---|
| All marl | 1 | 2 |
| All silt | 2 | 2 |
| All sand | 3 | 1 |
| Marl / Gravel Mix | 4 | 1 |
| All gravel | 5 | 2 |
| Fine aggregated marl | 6 | 5 |
| Agg. Marl / Pebble | 7 | 2 |
| All course agg. Marl | 8 | 3 |

For grab samples and combined net and pump samples, no significant correlation was found between sediment type and richness or abundance (table 9 and table 10 respectively).

Comparison of three sediment groups showed no significant difference in abundance or richness existed for grab samples (ANOVA $F_{2, 11}$= 1.8066, p≤ 0.2097 and $F_{2, 11}$ = 0.8732, p≤ 0.4447), for richness and abundance respectively. The same analysis for *C. luctuosa* and *A. aquaticus* also showed no significant difference (ANOVA $F_{2, 7}$ = 3.6672 p≤ 0.1280 and $F_{2, 7}$ = 0.2019 p≤ 0.6764) for *C. luctuosa* and *A. aquaticus* respectively.

Two-way ANOVA showed no significant relationship for the combined effect of depth and sediment type on total abundance and richness ($F_{2, 7}$ = 0.6635 p≤ 0.5447 and $F_{2, 7}$ = 0.4788p≤ 0.6384) respectively. The same analysis showed no significant relationship for *C.luctuosa* or *A.aquaticus* data from the fourteen grab samples ($F_{2, 11}$= 1.4379, p≤ 0.2792 and $F_{2, 11}$ = 0.6947, p≤ 0.5198) respectively

### 3.4 Variation in Community Structure among Sites

#### Grab Samples

Hierarchical clustering for grab samples grouped the two deep water sites together, i.e. they were more similar to each other than to any other sites. For the remaining sites, similarities in biotic communities among sites appeared to occur independently of water depth. Site 15 (depth 1.3 m) was the only obvious outlier (Figure 4).

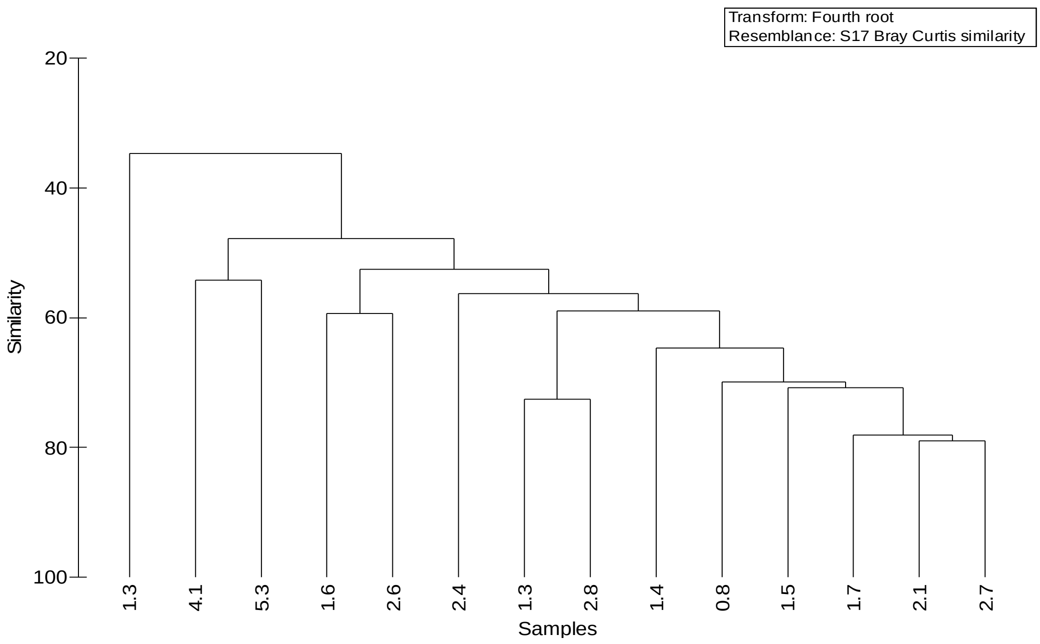


Figure 4. Hierarchical clustering of biotic data from grab samples using group average on Bray-Curtis similarity matrix. Samples labelled with depth

The MDS ordination for grab samples shows a similar pattern to the hierarchical clustering, with sites 3, 5 and 11 closely grouped and site 15 an obvious outlier. The circles on the ordination show sites that have been classified as > 60% similar by hierarchical clustering (Figure 5). The groupings determined by hierarchical clustering match those of the MDS reasonably well and this is indicative of an adequate MDS ordination.

Figure 5 has a stress value of 0.13 which can lead to some values being poorly represented on a 2 dimensional scale but overall this can still be considered a useful ordination.

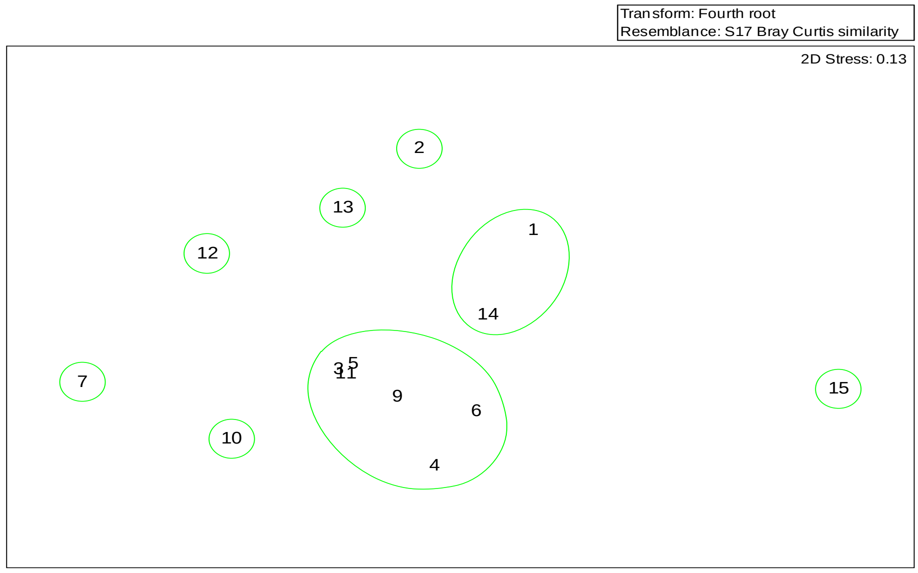


Figure 5. MDS ordination of biotic data from grab samples, overlain with 60% similarity as defined by hierarchical clustering. Stress 0.13

## Multivariate Analysis of Combined Net and Pump Samples

Hierarchical clustering of the net and pump samples did not appear to be influenced by depth but the depth range for these samples was narrow. Site 18 is classed as an outlier but overall no two sites could be considered strongly similar.

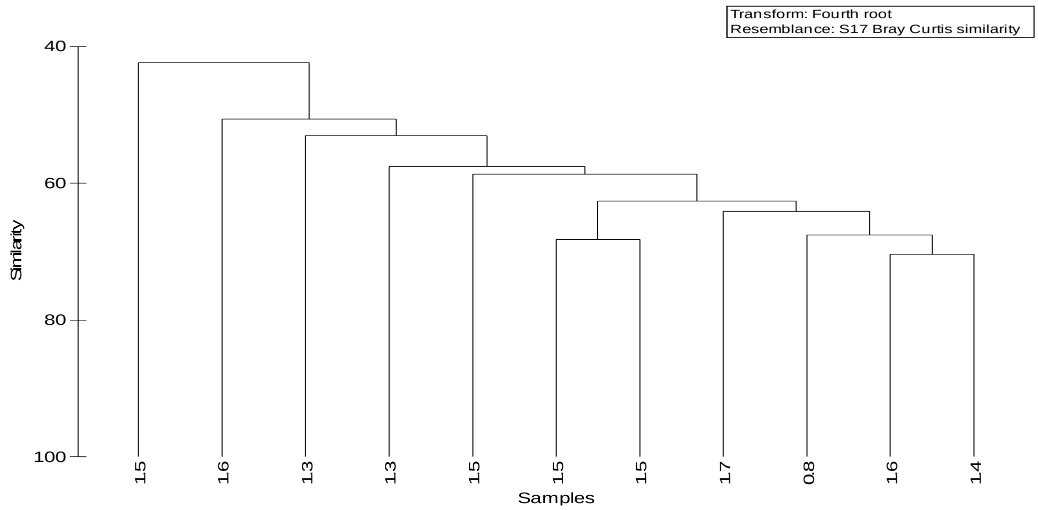


Figure 6. Hierarchical clustering of biotic data from combined net and pump using group average on Bray-Curtis similarity matrix. Samples labelled with depth

MDS shows a similar pattern to clustering, with no sites closely linked and site 18 shown as an outlier. Overlay of 60% similarity clusters corresponds reasonably well to the MDS groupings (Figure 8). The stress value of 0.12 shows that a degree of caution is necessary when analysing the results but the ordination is still a reasonable interpretation of the similarity matrix.

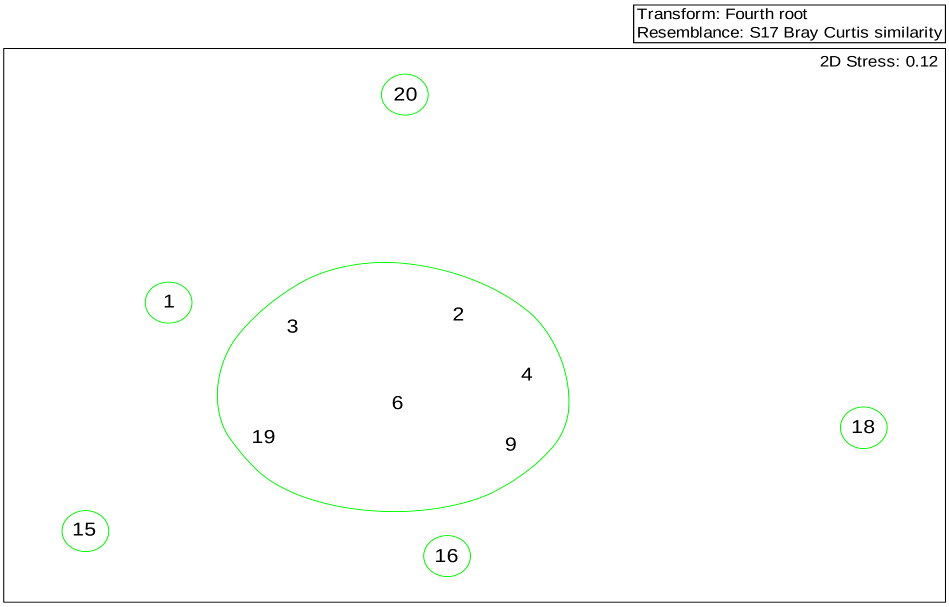


Figure 7. MDS ordination of biotic data from combined net and pump samples, overlain with 60% similarity as defined by hierarchical clustering. Stress 0.12

### 3.5 Sediment Chemistry

Chemical analysis results were listed in order of increasing depth (Table 6.). There was a marked difference in TP and Fe at the deep water sites (sites 7 and 12) in comparison to the mid-depth and shallow areas. The values for total nitrogen, manganese and magnesium were higher at sites 7 and 12 than elsewhere, although the difference was not as pronounced as for iron and total phosphorus. Total organic carbon was high at the deep water sites but a similar concentration was recorded in one shallow water area (site 20, 1.6 m). Calcium values varied widely across the lake, with low values at deep water areas but also in some shallow areas. The very low value for calcium at site 14 may be owing to an instrument error, as two replicates were tested for this sample and gave significantly different results (Appendix 2). The molar ratio of Fe to TP varied greatly across sites with depth showing no obvious influence. The values close to one are indicative of sediment where almost all iron molecules are bound to phosphorus (i.e. the sediment is near P saturation)

Table 6. Sediment chemistry, water depth and sediment classification results for all sites given in order of increasing depth. TOC=Total Organic Carbon % of dry sediment weight, TN= Total Nitrogen % of dry sediment weight, TP= Total Phosphorus, Fe= Iron, Mn= Manganese, Mg= Magnesium, Ca= Calcium, Fe:TP = Molar ratio of Fe to TP. (See table 1. for sediment classification)

| Site | Depth (m) | T N % | T P (mg $g^{-1}$) | Fe (mg $g^{-1}$) | Mg (mg $g^{-1}$) | Mn (mg $g^{-1}$) | TOC % | Ca (mg $g^{-1}$) | Sediment | Fe:TP |
|---|---|---|---|---|---|---|---|---|---|---|
| **6** | 0.8 | 0.34 | 0.044 | 0.292 | 0.938 | 0.053 | 9.01 | 412.72 | 7 | 3.7 |
| **1** | 1.3 | 0.40 | 0.136 | 0.701 | 1.074 | 0.122 | 9.23 | 404.81 | 4 | 2.9 |
| **15** | 1.3 | 0.38 | 0.022 | 0.056 | 1.120 | 0.240 | 8.01 | 394.71 | 1 | 1.4 |
| **9** | 1.4 | 0.32 | 0.036 | 0.511 | 0.856 | 0.073 | 8.86 | 419.57 | 5 | 7.9 |
| **18** | 1.4 | 0.63 | 0.086 | 0.117 | 1.245 | 0.679 | 9.91 | 451.72 | 6 | 0.8 |
| **4** | 1.5 | 0.25 | 0.038 | 0.259 | 0.751 | 0.050 | 8.65 | 365.50 | 8 | 3.8 |
| **16** | 1.5 | 0.47 | 0.043 | 0.053 | 1.203 | 0.301 | 8.73 | 429.37 | 6 | 0.7 |
| **19** | 1.5 | 0.63 | 0.093 | 0.213 | 1.126 | 0.710 | 10.52 | 397.04 | 6 | 1.3 |
| **2** | 1.6 | 0.41 | 0.066 | 0.412 | 0.951 | 0.077 | 10.95 | 404.45 | 7 | 3.5 |
| **3** | 1.6 | 0.28 | 0.054 | 0.655 | 0.883 | 0.101 | 8.69 | 428.32 | 5 | 6.7 |
| **20** | 1.6 | 0.66 | 0.085 | 0.159 | 1.299 | 0.884 | 11.17 | 387.63 | 6 | 1.0 |
| **5** | 2.1 | 0.39 | 0.076 | 0.575 | 0.977 | 0.095 | 9.85 | 419.04 | 8 | 4.2 |
| **10** | 2.4 | 0.41 | 0.089 | 0.432 | 1.039 | 0.071 | 9.75 | 409.58 | 8 | 2.7 |
| **13** | 2.6 | 0.42 | 0.140 | 0.765 | 1.045 | 0.118 | 9.51 | 368.41 | 6 | 3.0 |
| **11** | 2.7 | 0.37 | 0.102 | 0.931 | 0.869 | 0.126 | 9.23 | 396.62 | 3 | 5.1 |
| **14** | 2.8 | 0.30 | 0.097 | 0.430 | 1.059 | 0.066 | 7.65 | 303.96 | 1 | 2.5 |
| **7** | 4.1 | 0.67 | 0.539 | 2.343 | 1.511 | 0.166 | 11.12 | 368.10 | 2 | 2.4 |
| **12** | 5.3 | 0.69 | 0.545 | 2.280 | 1.442 | 0.159 | 11.51 | 355.81 | 2 | 2.3 |

The results of the current study were compared with chemical data from Lough Carra sediment samples taken in 2002 (Hobbs *et al.,* 2005). Accurate comparison of results is limited by the difference in mean depth at which the 2002 samples were taken (Table 6). The greatest similarity in mean depth between the two studies is for the south basin of the current study and the mid basin of the previous work. Comparison of these results shows higher mean TP today and lower values for all other variables. The mean Fe:TP ratio is far lower today for both basins which suggests a reduction in the nutrient binding capacity of the sediment since 2002.

Table 7. Comparison of chemical analysis (mean ± 95% CI) from this study with that of Hobbs *et al.,* 2005. TP = total phosphorus, Fe = iron, Mg = magnesium, Mn = manganese, %TOC = total organic carbon % of dry weight, Ca = Calcium, Fe : TP = Molar ratio of Fe to TP

| | Current Study | | Hobbs *et al.,* 2005 | | |
|---|---|---|---|---|---|
| | mid basin (n = 5) | south basin (n = 13) | north basin (n = 9) | mid basin (n = 7) | south basin (n = 7) |
| T P (mg $g^{-1}$) | 0.12 ± 0.06 | 0.15 ± 0.10 | 0.29 ± 0.06 | 0.09 ± 0.04 | 0.18 ± 0.08 |
| Fe (mg $g^{-1}$) | 0.56 ± 0.24 | 0.81 ± 0.38 | 3.31 ± 0.80 | 0.84 ± 0.45 | 1.76 ± 0.57 |
| Mg (mg $g^{-1}$) | 1.20 ± 0.07 | 1.03 ± 0.12 | 1.93 ± 0.18 | 1.68 ± 0.12 | 1.57 ± 0.34 |
| Mn (mg $g^{-1}$) | 0.07 ± 0.03 | 0.1 ± 0.02 | 0.11 ± 0.01 | 0.05 ± 0.01 | 0.1 ± 0.02 |
| % TOC | 9.67 ± 1.13 | 9.54 ± 0.60 | 9.3 ± 0.7 | 8.7 ± 1.0 | 8.7 ± 0.5 |
| Ca (mg $g^{-1}$) | 412.1 ± 24.0 | 389 ± 19.0 | 264.3 ± 23.0 | 257.1 ± 23.5 | 292.4 ± 19.4 |
| Fe : TP | 1.0 ± 0.27 | 3.9 ± 0.94 | 6.4 ± 0.7 | 5.6 ± 2.0 | 6.3 ± 1.7 |
| Depth (m) | 1.5 ± 0.10 | 2.3 ± 0.67 | 11.69 ± 3.10 | 4.4 ± 3.52 | 7.21 ± 2.97 |

**Correlation Analysis of Chemical Data**

Strong correlations were found between Log TP and each of the following: logFe, logMg, logTN, Mn. logTN and TOC were also strongly correlated with logFe and logMg. As was expected from examination of the raw data, depth was strongly correlated to logTP, logFe, logMg and Mn (Table 7).

Table 8. Pearson Product Moment Correlation Coefficients for abiotic data from all sites sampled. (n=18) *p<0.05 **p<0.001

| | Depth | TOC | LogTP | LogTN | LogFe | Ca | Sediment | Mn | LogMg |
|---|---|---|---|---|---|---|---|---|---|
| Depth | 1.000 | | | | | | | | |
| TOC | 0.458 | 1.000 | | | | | | | |
| LogTP | 0.768** | 0.709** | 1.000 | | | | | | |
| LogTN | 0.415 | 0.794** | 0.798** | 1.000 | | | | | |
| LogFe | 0.768** | 0.701** | 0.887** | 0.655** | 1.000 | | | | |
| Ca | -0.559* | 0.044 | -0.377 | 0.061 | -0.216 | 1.000 | | | |
| Sediment | -0.490 | 0.149 | -0.423 | -0.141 | -0.376 | 0.507 | 1.000 | | |
| Mn | 0.729** | 0.620** | 0.809** | 0.483* | 0.950** | -0.204 | -0.297 | 1.000 | |
| LogMg | 0.602** | 0.511* | 0.803** | 0.899** | 0.589 | 0.140 | -0.478* | 0.450 | 1.000 |

No significant relationship was found, between species richness or abundance, and any of the abiotic data (Tables 9 and 10). For combined net and pump samples *C. luctuosa* abundance was strongly correlated to total abundance, reflecting the dominant nature of this species at most shallow and mid-depth sites (Table 10). A similar correlation was not found for grab samples where some sites were over 4 m deep (Table 9).

Table 9. Pearson Product Moment Correlation Coefficients for grab samples. (n= 15). *p<0.05 **p<0.01

| | Abundance | Richness | *C. luctuosa* | *A. aquaticus* |
|---|---|---|---|---|
| Abundance | 1.000 | | | |
| Richness | 0.286 | 1.000 | | |
| *C.luctuosa* | 0.069 | -0.021 | 1.000 | |
| *A. aquaticus* | 0.299 | 0.434 | -0.332 | 1.000 |
| TOC | -0.108 | 0.270 | -0.377 | -0.003 |
| LogTN | -0.297 | -0.195 | -0.539 | -0.199 |
| LogTP | -0.178 | -0.309 | -0.595 | -0.272 |
| LogFe | -0.168 | -0.064 | -0.555 | -0.259 |
| logMg | -0.283 | -0.519 | -0.462 | -0.145 |
| Mn | -0.105 | 0.058 | -0.491 | 0.071 |
| Ca | -0.280 | 0.427 | 0.411 | -0.144 |
| Sediment | 0.346 | 0.550 | 0.272 | 0.280 |
| Depth | -0.037 | -0.238 | -0.717 | 0.020 |

Table 10. Pearson Product Moment Correlation Coefficients (biotic data for combined net and pump samples). (n= 11). *p<0.05 **p<0.01

| | Abundance | Richness | *C.luctuosa* | log *A.aquaticus* |
|---|---|---|---|---|
| Abundance | 1.000 | | | |
| Richness | 0.186 | 1.000 | | |
| *C.luctuosa* | 0.953** | 0.103 | 1.000 | |
| log *A.aquaticus* | 0.410 | 0.490 | 0.383 | 1.000 |
| Depth | -0.422 | 0.143 | 0.330 | -0.019 |
| Sediment | -0.025 | 0.018 | 0.079 | -0.046 |
| TOC | -0.332 | 0.294 | -0.357 | -0.070 |
| Ca | -0.182 | -0.189 | -0.239 | -0.115 |
| Mn | 0.165 | 0.365 | -0.031 | 0.413 |
| LogMg | -0.443 | -0.009 | -0.558 | -0.532 |
| LogFe | -0.167 | 0.440 | -0.329 | 0.057 |
| LogTP | -0.134 | 0.170 | -0.270 | -0.208 |
| LogTN | -0.505 | -0.043 | -0.563 | -0.580 |

### 3.6 Multivariate Analysis of Abiotic Data

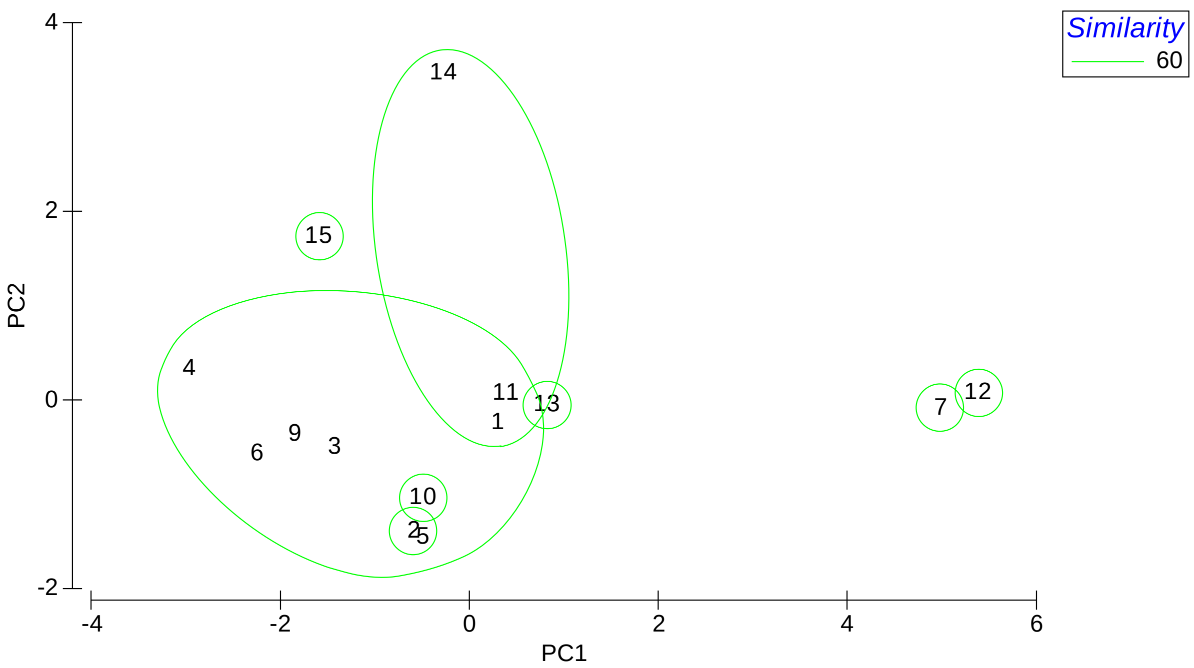


Figure 8. 2-dimensional PCA ordination of abiotic variables from all sites overlain with 60% similarity as defined by MDS of the biotic data.

The ordination highlights the marked difference of the two deep water sites (7, 12) to the other locations. Site 14, which has a very low calcium concentration, is also an outlier.

The ordination was visually inspected for similarities to the MDS ordinations generated for the biotic data. No strong similarities were obvious, e.g. samples 3, 5 and 10 are closely grouped according to abiotic data in the PCA but show little similarity in relation to biotic data in the MDS plot. Overlay of 60% similarity from MDS biotic grab data onto the PCA plot for chemical data shows considerable contradiction e.g. biotic data for sites 1, 11 and 14 are classed as 60% similar but PCA of the abiotic data for the same sites detects very little similarity among them.

### 3.7 Analysis of Combined Biotic Data and Abiotic Data

A visual comparison of the association between community composition and specific environmental variables was made using the MDS ordinations overlain with values for abiotic variables. No evidence of relationships was found (see Figure 9 and 10 for examples).

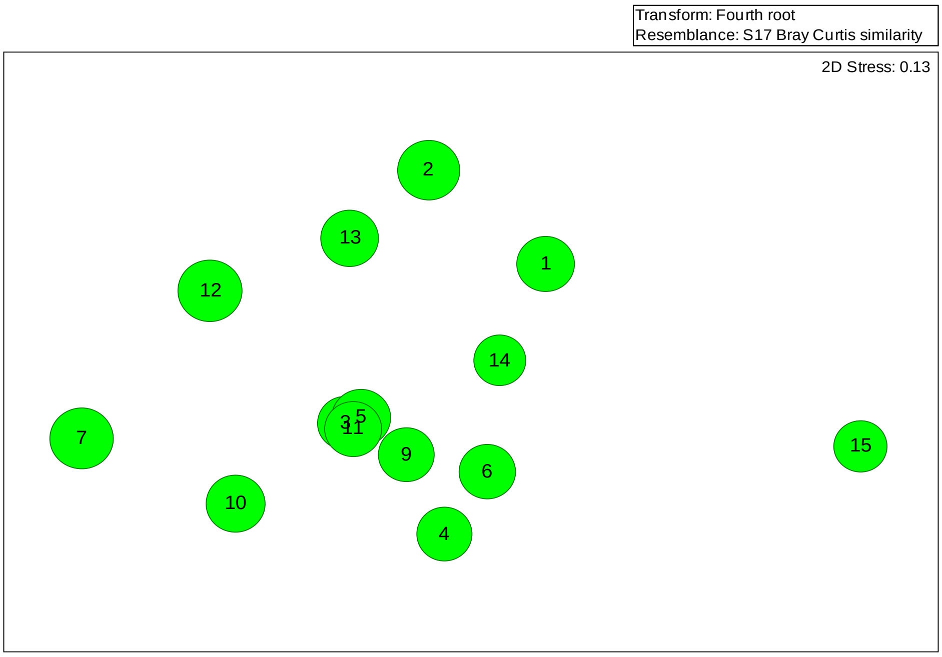


Figure 9. MDS ordination of biotic data from grab samples overlain with TOC values. Larger bubble indicates higher concentration of TOC.

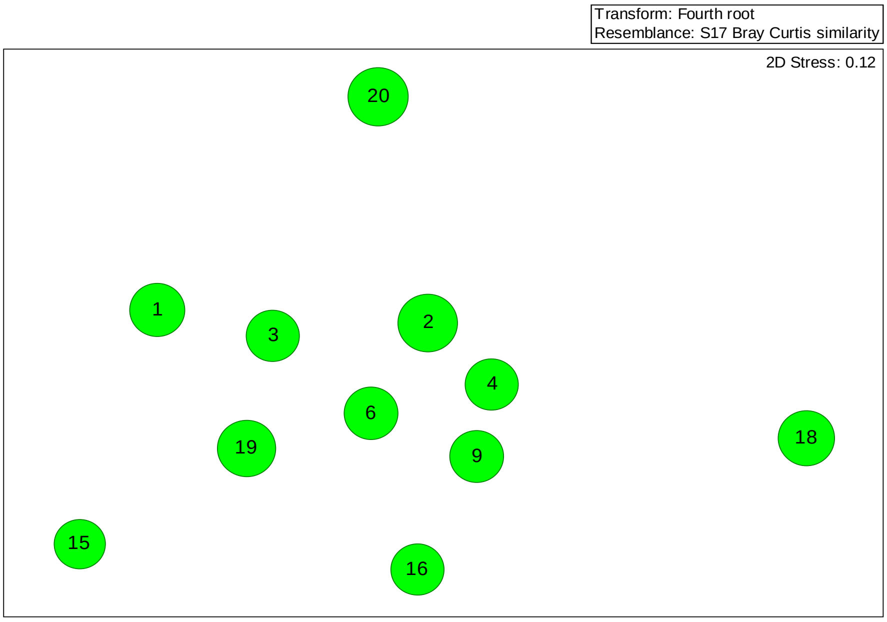


Figure 10. MDS ordination for biotic data from combined net and pump samples overlain with values for TOC. Larger bubble indicates higher TOC concentration.

The highest value generated for rank correlation between abiotic variables and biotic grab data by the BEST procedure was 0.517 for the combination of Mn, logMg and TOC. This low value suggests that that no combination of environmental variables was closely matched to the biotic data. The match between biotic and abiotic data for net and pump samples was lower again with a maximum Rho value for combined pump and net samples of 0.429, with Mn, logMg and Ca classed as having the highest correlation with the biotic data.

For combined net and pump samples, the Global Best test calculated a test significance of 26% at Rho 0.429, indicating that the level of similarity between abiotic and biotic data, calculated by the BEST procedure, was not significant. For grab samples, the Global Best test gave a test significance of 5% but the low value of the sample statistic (Rho 0.517), suggests no significant relationship was found between grab biota samples and environmental variables.

## 4. Discussion

### 4.1 Comparison of Sampling Methods

The most noticeable feature of the methods comparison is the low values for total abundance achieved with the grab compared with net or pump samples. This result is to be expected when the ecology of macroinvertebrates is considered. All macroinvertebrates require oxygen for respiration, albeit in varying amounts depending on species (Kolar and Rahel, 1993, Bachmann and Usseglio-Polatera, 1999, Chaves *et al.,* 2005). Oxygen concentrations in lake sediment generally fall with depth into the substrate (Meijer and Avnimelech, 1999, Matisoff and Neeson, 2005); as a result, the majority of macroinvertebrates will be found in the upper few centimetres of the sediment layer. The Ekman grab samples a relatively small surface area (225 $cm^2$),and in soft sediment the jaws can penetrate deeply (maximum grab volume 3500 $cm^2$). As a result a high a high portion of the sample will consist of deeper, low oxygen sediment. (Schloesser and Nalepa, 2002).

In contrast, the net sampling involved movement of the net mouth across the sediment surface for thirty seconds, thereby covering a larger surface area than the grab and sampling less of the deeper sediment. Pump sampling 27 litres of sediment/water mix, also covered a large surface area and removed only the upper, specimen-rich, sediment layer.

The similarity between results for net and pump samples is in agreement with previous work by in Lough Carra (McGoff and Irvine, 2009), and is not surprising when the sampling sites are considered. Both samplers work efficiently in the type of areas sampled in this study: soft sediment, free of large stones and macrophyte cover. It is primarily the upper layer of sediment which is sampled by both methods and so a yield of similar community composition is to be expected.

Despite the far higher abundance from net and pump samples, no significant difference in species richness was found among the three methods. If taxonomic richness rather than abundance values are required, grab samples may be as effective at the net and pump sampling. However, the number of sites available here for comparison is low and grab sampling did have the lowest mean species richness (Table 3).

Overall, net sampling was the most practical method for the shallow water sites. If areas with large rocks are to be sampled the pump may be a better choice as the tubing can be inserted between the rocks while the net would bounce over them. However, pump sampling is time consuming and laborious, involving at least two operators and the sieving of a large quantity of sediment / water mix. In contrast, net sampling can be swiftly performed by one person and the resulting sample is easily sieved. In light of the similar values obtained from the two methods, net sampling was the better option in this study.

When interpreting species richness values from any of these methods, it is worth noting that the samples taken may represent only a small number of species present in that environment. A recent study compared grab sampling to the Chironomid pupal exuvial technique as a method to estimate benthic macroinvertebrate community structure in lakes. This study found that the number of chironomid species detected with the exuvial technique was greater than the total number of invertebrate species found with grab sampling (Raunio *et al.,* 2007). Such research highlights what a limited representation of biotic communities may be shown by one sampling technique.

### 4.2 Sediment Chemistry

When compared with values for other Irish lakes surveyed by the Environmental Protection Agency (EPA), the sediment surface concentrations of TP and Fe in Lough Carra are very low. The EPA IN-SIGHT project involved analysis of over thirty oligotrophic and mesotrophic lakes (mean annual water column TP 0 – 17 µg $l^{-1}$), which were all considered as candidate reference lakes under the Water Framework Directive (2000/60/EC) i.e. free of, or subject to minimal, anthropogenic impact. For these lakes, surface sediment mean values for TP ranged from approximately 0.3 – 3.2 mg $g^{-1}$ and Fe values ranged from below 5 mg $g^{-1}$ to over 150 mg $g^{-1}$ (Taylor *et al.,* 2007). The samples for the IN-SIGHT project were taken from the deepest part of each lake and as this project has shown, sediment at deeper sites can contain far higher concentrations of TP and Fe than in shallow areas. However, even when compared with samples at similar depths, TP and Fe values for Lough Carra are similar to the lowest concentrations found among the thirty two lakes in the IN-SIGHT project.

It is worth noting that under the IN-SIGHT project, seven lakes which have been subject to considerable anthropogenic impact were also surveyed. Water chemistry values for TP in these lakes ranged from 19.3 – 675 µg $l^{-1}$ but TP concentrations in the sediment were similar to those found in oligotrophic lakes. Therefore, low TP in the sediment of Lough Carra is no guarantee that the lake is free from nutrient enrichment. It must also be noted that, although the chemical values were low in Lough Carra compared with other lakes, these chemicals may still be impacting on a lake ecosystem which was once considered pristine.

Most of the high correlations found among the chemical data in this project are in keeping with previous research regarding metal / nutrient interactions. It has been well-established that phosphate binds to manganese and iron under oxic conditions, to form precipitates (Jordan and Rippey, 2003, Katsev *et al.,* 2006). An oligo-mesotrophic lake such as Lough Carra should have a reasonably high level of oxygenation and the high correlation of logTP to logFe and Mn is therefore, expected, as is the correlation of logFe to Mn. Previous studies have shown that phosphorus also precipitates with calcium (Dittrich and Koschel, 2002, Virginia *et al.,* 2006) but no correlation was found between these elements. It is likely that depth and metal binding have a stronger influence on TP concentrations than calcium.

Lower oxygen concentrations in deeper water generally lead to reduced binding of Fe to P. The high concentrations of TP, Fe and Mn at deep sites could be taken as an indication of depth exerting the opposite effect on nutrient/metal binding i.e. a positive one. However, the molar ratio of Fe:TP ratio varies greatly across sample sites, independently of depth e.g. the low ratio of site 15 suggests it is nearing saturation while the high ratio at site 9 suggests abundant free iron molecules. These two sites were sampled at a very similar depth. This suggests that the sediment in deep areas does not have a greater capacity to bind nutrients than sediment in shallow areas. It is more likely that a reduction in $CaCO_3$ precipitation accounts for the high values of TP, Fe and Mn at deep sites.

Visual examination of the data set reveals the strong influence of depth on sediment chemistry and this is confirmed by the correlation calculations. The negative correlation of Ca and depth can be explained by the influence of photosynthesis. Shallow water sites have higher rates of photosynthesis than deep sites, which results in a higher rate of $CO_2$ removal from the water and therefore, higher pH. This increase in pH causes precipitation of $CaCO_3$ (Heath *et al.,* 1995), leading to build-up in the lake sediment.

This lower rate of $CaCO_3$ accumulation in deep areas may be partly responsible for the high metal concentrations at these sites. If the accumulation rate of other elements is not altered at deep sites, their concentration in the sediment will be higher owing to less dilution with $CaCO_3$. The fact that concentrations of Fe, Mg, TP, TN and TOC are higher at deep sites, while $CaCO_3$ concentration is lower, supports this theory of a reduced accumulation rate in one element. Previous studies have shown that varying accumulation rates can play a major role in determining lake sediment chemistry (Engstrom and Wright, 1984). This dilution does not fully explain the differences in metal concentration, as some shallow sites have low Ca and low metal concentrations. This may be owing to higher bioturbation rates in shallow areas which can stimulate the flux of metals from the sediment to the water column (Krantzberg, 1985).

The higher TOC and TP values in the deep areas may also be partially explained by reduced $CaCO_3$ accumulation but slower decomposition would also play a part. Organic matter from the water column settles on the sediment surface throughout the lake, but at deep water sites, the low light and oxygen levels result in slow decomposition of this matter (Muller *et al.,* 2003).

Chemical analysis of surface and core sediment in Lough Carra was carried out on samples taken in 2002 (Hobbs *et al.,* 2005). This work showed significant differences in sediment chemistry among the three basins of Lough Carra. Direct comparison of the current project with the 2005 work is limited by the difference in mean depth at which samples were taken (Table 7). The greatest similarity in mean depth between the two studies is for mid-basin samples from 2005 (mean depth 4.4 m) and south basin samples in the current project (mean depth 2.3 m).

Comparison of these two data sets show higher Ca values today which may be explained by the shallower depth of the sample sites. However, higher TOC and TP levels were recorded in the current study. This may be a result of continuing or increasing nutrient loading into the lake. Of particular interest is the fact that the Fe:TP molar ratios are much lower in the current study which suggests considerable reduction in the sediment binding capacity since 2002. The mid-basin in particular, appears to have very little nutrient-binding capacity remaining. However, in the current study Fe:P varied greatly across sites, from areas which appear saturated to sites with abundant binding capacity remaining. It must also be noted that the Fe:TP ratio can only be taken as an estimate of sediment binding capacity. The exact portion of total phosphorus bound to iron and other molecules in the sediment is unknown. A more extensive study of the lake, including analysis of bound and unbound phosphorus concentrations, would be required to determine if a reduction in binding capacity has occurred across the lake.

The study of Hobbs *et al.,* concluded that sediment chemistry heterogeneity among the basins was largely the result of riverine inputs. However, in that study, the depth of sample sites varied considerably among three basins. The mean levels of TN, TP, Fe, Mn and TOC increased with greater mean depth of sample sites i.e. lowest values were in mid basin which had the shallowest mean site depth, highest values were in north basin which had the highest mean depth. This concurs with the finding of the current study, regarding the strong influence of depth on sediment chemistry. However, $CaCO_3$ concentrations do not show a corresponding decrease with depth across the three basins. The lowest concentration of Ca was found in the south basin where the mean depth of sample sites (7.21 m) was deeper than that of the mid basin (mean depth 4.40 m). It may the case that the higher water column TP, recorded by Hobbs *et al.,* in the north and south basins, lead to higher rates of photosynthesis which generated increased $CaCO_3$ precipitation in these basins.

The high levels of TP and low Fe:TP ratio in deep water sediment poses a serious risk to the ecological integrity of the lake. The results of this and several other studies suggest that nutrient input to Lough Carra has increased in recent years. If this input continues to increase, or remains constant, all buffering capacity of the lake may soon be lost. At present, phosphorus input may be far higher than is apparent from examination of biomass in the water column because the lake sediment is still rendering much of the P unavailable to organisms. If P saturation of the sediment is reached, a far higher percentage of P entering the lake would remain bioavailable. A rapid change in trophic status is the likely outcome. The subsequent reduction in oxygen levels would cause areas which are currently low in oxygen (i.e. deep water sites which are high in TP) to become anoxic. Anoxic conditions would lead to the reduction of Fe and Mn in the sediment and release of phosphate to the water column, causing more severe eutrophication.

Such a natural-feedback mechanism would be very difficult to halt or reverse. Preventing the occurrence of sediment saturation by, improving landuse practises in the catchment and thereby reducing nutrient input to the lake, is a far more practical and ecologically sound option.

## 4.3 Macroinvertebrate Distribution

### 4.3.1 Biotic Data Overview

The most prominent feature of the biotic data set; the wide distribution and high abundance of *Caenis luctuosa*, can be partially attributed to environmental conditions within the lake. *C.luctuosa* feeds by collecting fine particulate organic matter and so is not restricted to areas with abundant periphyton cover or course organic matter (Elexová and Némethová, 2003) Members of the genus are known to be pollution-tolerant and so should not be greatly affected by nutrient enrichment (Landa and Soldan, 1985). Another important factor to consider is the type of sampling sites chosen for this study. Only sites free of macrophyte growth were sampled. Previous work in the same lake found a negative association between *C.luctuosa* and macrophyte cover (McGoff and Irvine, 2009).

The lifecycle of *C.luctuosa* also plays a part in explaining why such high numbers were found. In temperate climates, *C. luctuosa* usually display a univoltine or divoltine lifecycle (Gonzalez *et al.,* 2001). The emergence of sub-imago Ephemeroptera often occurs over a very brief, concentrated, period, with huge numbers of insects emerging from one location, in the space of a few days. Mating of the adults and laying of eggs is also concentrated to a short time-span (Harker, 1992, Corkum *et al.,* 2006). This highly co-ordinated lifecycle can result in large numbers of eggs hatching and maturing at the same time. Sampling for this project took place in late April, and the large-scale emergence of Ephmeroptera species usually occurs from May onwards. It is likely that when sampling took place, the *Caenis* nymphs were nearing their maximum population density before a mass emergence took place. Previous work on Lough Carra confirms that significant seasonal variation of *Caenis* spp. occurs within the lake. Little (2008) found that total abundance of *Caenis* spp. dropped dramatically from spring to summer and attributed this to the univoltine lifecycle of the genus in that region.

The tolerance of *Caenis* spp. to a broad range of environmental variables may explain its wide distribution within the lake and its lifecycle explains seasonal variation in abundance. However, the exact reason for such high numbers within the lake is not clear. Little (2008) surveyed nine lakes, with high, mid and low alkalinity each represented by three lakes. *Caenis* spp. were the most abundant invertebrate in two high alkalinity lakes but also in one low alkalinity lake. Other lakes across the alkalinity range showed low *Caenis* abundance. The TP concentration of these lakes was not an obvious influence on the Caenid abundance, although the mean TP range across the lakes was relatively narrow ($5 – 15\ \mu g\ l^{-1}$). The lake with the highest TP and that with the lowest both showed low Caenid abundance. However, work by the EPA on over fifty lakes (including Lough Carra), with a similar TP range to the lakes used by Little, showed a strong association between *Caenis* spp. and high alkalinity lakes (Free *et al.,* 2006). It is clear that alkalinity does play a role in the dominance of *C. luctuosa* in Lough Carra but in light of the variability of the species in other high alkalinity lakes, other environmental factors must also play a part.

The broad distribution of *A. aquaticus* and Chironomidae spp. is to be expected as these taxa can tolerate a wide range of environmental conditions, are not fastidious feeders (Rosenberg and Resh, 1993) and are very common taxa in Irish lakes.

The presence of *Crangonyx pseudogracilis* in the lake is worthy of mention. This is the first record of the invasive North American species in Lough Carra. *C. pseudogracilis* has been recorded in Ireland since 1969 but by 1993 its' expansion range was still considered very narrow (Costello, 1993). Laboratory experiments and extensive field work in Lough Neagh have shown that although *C. pseudogracilis* can tolerate the same physio-chemical conditions as Gammarus species, it suffers from predation by *Gammarus duebeni* and

*Gammarus pulex*. As a result *C. pseudogracilis* rarely occurs in the same micro-habitat as the gammarus species (Dick, 1996, MacNeil *et al.,* 2000, MacNeil *et al.,* 2003a). However, *G. duebeni* is commonly infected with a microsporidian parasite, *Pleistophora* sp. which does not infect *C. pseudogracilis*. Infection with this parasite reduces the ability of *G. duebeni* to prey on *C. pseudogracilis* and could potentially lead to the dominance of *C. pseudogracilis* within a habitat (MacNeil *et al.,* 2003b).

Amphipod data from this project is in keeping with previous research regarding co-habitation of species. *G. duebeni* and *G. pulex* numbers are very low, with only three specimens of each species found. In contrast, *C. pseudogracilis* was the most dominant amphipod present (88 specimens across 5 sites). It appears that the low numbers of *G. duebeni* and *G. pulex* have allowed the spread of *C. pseudogracilis*. Interestingly, the native Gammarid *G. lacustris* was relatively widespread in the lake (27 individuals across 5 sites) and was found at four of the sites where *G. lacustris* was present. Interspecies predation does not appear to occur between these to species.

The impact of the *C. pseudogracilis* on the ecology of Lough Carra is, as yet, unknown. Since predation by the native *G. lacustris* does not appear to occur and other Gammarid numbers are very low, the potential exists for the alien species to dominate the amphipod community within the lake, leading to further reduction in Ireland's native fauna. This may have consequences for species on trophic levels both above and below amphipods.

Low nutrient, hard-water lakes such as Lough Carra, generally provide a good environment for gastropods and the low numbers here are probably a reflection on the absence of macrophyte cover at the sampling sites; grazers such as gastropods require macrophytes to provide substrate for periphyton (Bronmark and Hansson, 2005).

The low diversity of Ephemeroptera and in particular, *Ephemera danica*, is worthy of mention. *E. danica* is of great importance to the Irish fly-fishing industry as the mass emergence of the sub-imago in May and June provide ideal conditions to catch salmon and brown trout by fly-fishing. *E danica* is a widely dispersed species in Ireland and commonly inhabits the soft sediment of high alkalinity lakes (Kelly-Quinn and Bracken, 2000). Lough Carra meets these conditions but the species was found at just two sites (13 specimens at site 3, 2 specimens at site 5). The only distinguishing feature of these sites was very high Ca concentration, a more detailed survey could reveal if this is an important influence on *E. danica* distribution.

Low *E. danica* abundance in Lough Carra has also been recorded by Little (2008) and McGoff (2009). Local anglers have, in recent years, reported a major decline in *E. danica* numbers in Lough Carra and a subsequent reduction in catches of brown trout. *E. danica* is pollution-sensitive and this decline is most likely owing to nutrient enrichment of the lake, with intensive agriculture in the catchment as the probable source. A definitive statement cannot be made on the decline of *E. danica* in Lough Carra as no historical records exist for its' level of abundance within the lake. However, as a location which was once nationally renowned as a brown trout fishery and considering the suitability of sediment and water hardness, it is reasonable to assume that Lough Carra supported a high population of *E. danica* in the past.

The evidence suggests that one of Ireland's finest fly-fishing locations has been degraded by poor management and this could be very damaging to the country's reputation as a world-class angling destination.

### 4.3.2 Effect of Depth on Biota

Of all the environmental variables measured, it was depth which demonstrated the most pronounced effect on macroinvertebrate community structure. However, it was at the deep water sites (4 m+) only, where depth had an obvious effect. In these areas the community structure was altered significantly. The species common in shallow regions, *C.luctuosa* and *A.aquaticus* were almost completely absent, and the areas were dominated by Chironomidae and Sphaeriidae, species adapted to cope with low oxygen conditions.

Despite the obvious changes in community structure noted by visual examination of the raw data, correlation analysis showed no significant relationship between depth and taxa richness or abundance. In deep water sites, where competing species are excluded, the remaining successful species can reach high abundance levels. However a drop in species richness with depth is to be expected. The absence of this relationship is partially owing to the low number of deep water sites sampled (only two), and so their influence on the complete data set is not very strong. The shallow and mid-water sites show high variation in richness and abundance, independently of depth. It appears the change in depth between shallow and mid-water sites does not cause dissolved oxygen concentration to decrease to a point where some species are excluded and so richness may not be greatly altered. It is possible that depth affects richness across the shallow to mid-water range but the effect is a subtle one and a greater number of samples are required to demonstrate this effect. The maximum number of samples available for comparison between mid and shallow sites was twelve for grab samples, and only seven for combined net and pump. Any outliers in these sample sets would skew the results of correlation analysis greatly.

In contrast to overall species richness, *C. luctuosa* numbers did drop relatively consistently with depth, in shallow, mid and deep water areas and as a result, show a strong negative correlation with depth. It is likely that overall taxa richness does not follow the same pattern as *C.luctuosa* because the decrease in oxygen between shallow and mid-water sites may not be enough to exclude a species completely, although it can limit their ability to feed and reproduce, leading to lower numbers. This appears to be the case with *C. luctuosa*.

It is clear from this study that despite the relatively shallow nature of Lough Carra, depth exerts a strong influence on the benthic invertebrate community. This influence must be considered not only when comparing biotic data from Lough Carra with that of other lakes, but also when comparing sites within Lough Carra.

### 4.3.3 Effect of Substrate Type on Biota

Despite the wide variety of sediment types sampled within the lake, no obvious pattern was evident, between macroinvertebrate distribution and sediment type. Reports in the literature vary regarding the sediment preference of the most dominant species found here, *C. luctuosa*. A study on the River Danube found higher *C. luctuosa* numbers in sand/gravel sediment compared with finer mud substrate (Elexová and Némethová, 2003). In contrast, White and Irvine (2003) surveyed 15 different mesohabitats within one lake and found no *C. luctuosa* in sand/marl sediment.

In the current study, high *C. luctuosa* abundance was found across a range of sediment types, from very fine marl to gravel and course aggregated marl. This suggests the species can adapt to a variety of sediment conditions. Reports of the species burrowing in, or clinging to, the sediment, adds weight to this theory. It may be the case that the insect will burrow when sediment is soft enough but can cling to the surface of harder substrate.

At many of the sampling sites, the sediment was not well-sorted, possibly as a result of sheltered conditions and associated limited wave action. This poorly sorted sediment may explain the lack of response in community composition to changing sediment type. Sediment was classified according to increasing dominance of large particles but many sites contained a mix of fine marl or sand and course pebble or aggregated marl. Changes in the relative composition of large and small particles among these sites may give different classifications, but the suitability of the substrate for different species may be unchanged as enough fine and course material may still be present to suit each taxa.

Many studies on rivers have found distinct communities in different sediment types (Rieradevall *et al.,* 1999, Heino, 2000, Cascorbi, 2002, Graca *et al.,* 2004). In such cases, the environmental conditions associated with specific sediment types may also influence the community composition. For example, distinct communities may be found in lotic sites with course, gravel-like sediment. This sediment is associated with high flow, erosional areas, which are often well-oxygenated and contain abundant course particulate organic matter (Elexová and Némethová, 2003). These environmental conditions can have a strong influence on macroinvertebrate community composition. In contrast to rivers, lakes may not have such distinct environmental conditions associated with a particular sediment type and so the community composition may be more independent of substrate. In a calcareous lake, such as Lough Carra, fine-grained substrate may result from $CaCO_3$ precipitation induced by high photosynthesis but in a river, fine sediment usually results from decreased flow rate.

Previous work by White and Irvine (2003) on a calcareous mesotrophic lake did, however find that distinct macroinvertebrate communities were associated with sediment of a specific particle size. The difference in sampling regime between this project and that of White and Irvine, may explain the conflicting results. In the 2003 work five replicate samples were taken in each mesohabitat of the lake, using one sampling technique (net) and all samples were taken at a depth of less than 0.5 metres. The broad objectives and limited time of this current study, did not allow for such replication. At each site, only one sample was taken with each method, or two in the case of grab samples but these samples were then combined. The samples were taken over a broad range of depths and some sediment types occurred at very few sites. The end result of all this variability was that some sediment types were represented at two sites or less, these sites may have been sampled at a different depth and with only one sample taken at each. Even when sites were grouped into broader sediment categories, no influence on community structure was obvious, but broader sediment categories resulted in a wide range of sample depths within each sediment category.

When the results are considered in the context of related research, it is likely that sediment type does exert some influence on macroinvertebrate community structure in Lough Carra, but this influence was obscured by natural variability among sites and a lack of sample replicates.

### 4.3.4 Influence of Chemical Variables on Macroinvertebrate Distribution

The chemical variables measured did not show any clear relationship to the invertebrate community structure observed at each site. PCA ordination of chemical data and the MDS analysis for the biotic data, produced very different results. Sites placed close together on the MDS showed little similarity in the PCA.

For some of the chemical variables measured, (Fe, Mn, Mg), a direct relationship with community composition is not to be expected, unless they are present in such high concentrations as to be toxic to organisms. Primary producers have such low requirements for these elements that they are rarely limiting to growth (Bronmark and Hansson, 2005). Low levels of variance in concentrations of these chemicals would not be expected to affect rates of photosynthesis and dissolved oxygen levels in the water. Therefore, macroinvertebrate communities should be unaffected by minor changes in these elements. The measurement of these chemicals was intended to shed light on how they interact with each other and with nutrients in the sediment, rather than their direct effect on fauna.

Macroinvertebrates have an obvious dependence on organic carbon as this is the main constituent of their food intake. This dependence was not apparent in the data generated here, where macroinvertebrate abundance and richness varied independently of TOC concentration. Previous studies have shown that high levels of organic matter can lead to reduced species richness owing to higher primary productivity and lower oxygen levels. Alternatively, very low concentration of organic matter results in limited food supply and lower abundance and diversity (Takamura *et al.,* 2009, Miyake, 2002). It appears that the TOC content in the lake did not fall into either of these extreme ranges, therefore a pronounced effect on the biotic community was not observed.

In the case of TP, the issue of bioavailability discussed earlier must also be considered. The high levels of calcium carbonate may have bound a significant portion of the total P present and thus render it unavailable to primary producers (Dittrich and Koschel, 2002). Therefore the level of TP recorded at each site may be a poor reflection on the amount influencing primary productivity rates.

When attempting to explain the effect of nutrients and oxygen concentration on a macroinvertebrate community, the tolerances of species within that community must be considered. This may offer the best explanation for why no relationship was found between nutrients and community composition. The four most widespread and abundant taxa found across the sample sites have a tolerance that stretches well above and below, the trophic conditions found at Lough Carra, where mean TP was 10.4 µg $l^{-1}$ (±1.3 µg $l^{-1}$) in 2003 (Irvine *et al.,* 2003). Of all the taxa recorded at the site, only *Gammarus lacustris* is primarily associated with low nutrient conditions (Rosenberg and Resh, 1993, O'Toole *et al.,* 2007).

Therefore, a high degree of spatial variation in nutrient level within the lake may be necessary before a change could be detected in the invertebrate community.

The lack of species associated with oligotrophic conditions, within Lough Carra, may be a warning sign of a decline in ecological quality of the lake. As a candidate reference lake under the Water Framework Directive (Directive 2000/60/EC), Lough Carra would be expected to support a community of pollution-intolerant species. The absence of such a community casts doubt over the EPA classification of Lough Carra as having good ecological status. However, it must be borne in mind that only a limited range of habitats were surveyed within the lake for this study.

#### 4.3.5 Further Study

The results of this project, and comparison with previous work, show a strong relationship between sediment chemistry and depth in Lough Carra. A wide range of interactions may explain the relationship and the potential exists for extensive further research in this area. Replicate sediment samples could be taken at depths across the entire bathymetric range of the lake and values for nutrients and metals compared. Such replication was lacking in this project and limited the comparisons which could be made. Of particular interest would be further investigation of $CaCO_3$ precipitation and its' influence of chemical concentrations. Sediment cores could be aged and the accumulation rate of $CaCO_3$ and other chemicals estimated. Such research could determine if higher metal and nutrient concentrations are the result of reduced $CaCO_3$ precipitation.

This work could be expanded to investigate the cause of $CaCO_3$ precipitation. Chlorophyll-α concentration in the water column could be compared with $CaCO_3$ levels in the surface sediment, to investigate the relationship between the rate of photosynthesis and $CaCO_3$ precipitation. Evidence of a relationship between primary productivity and $CaCO_3$ accumulation could be examined over a longer time period using palaeolimnological evidence from sediment cores. This work would aid attempts to determine the source and rate of nutrient loading in the sediment of Lough Carra.

The potential exists for further work on the relationship between macroinvertebrate and nutrients. In this study, the only concentration of phosphorus measured was total phosphorus. The concentration of bioavailable phosphorus which is loosely bound to, or present in the interstitial spaces of, sediment could be measured and compared with the macroinvertebrate community present. An extension of this work could involve more detailed analysis of the organic matter present in the sediment. Rather than measuring only total organic carbon, the form of the carbon present could be analysed i.e. living or dead periphyton and bacteria, course particulate organic matter. The relationship between particular forms of organic matter and the specific macroinvertebrate could be investigated. In particular the feeding behaviour of *C. luctuosa* within the lake could be examined with a view to explaining its' high abundance in the lake.

This study highlighted the wide variety of sediment types present in Lough Carra. Although no relationship was found between macroinvertebrate distribution and sediment, the high level of variability among sites and lack of replicates hindered analysis of such a relationship.

Distinct sediment types could be identified in the lake and a high number of replicates could be taken for each sediment type, within a narrow depth range. Such work could reveal subtle effects of sediment type on macroinvertebrate community composition.

The low numbers of *E. danica* should be investigated further. As their value to the angling industry demonstrates, they represent not just an ecological asset but an economic one also. A wide ranging survey of mesohabitats within the lake, combined with water and sediment chemical analysis, could determine the abundance of *E. danica* and the conditions which affect its' distribution.

## 5. Conclusions

Grab sampling yielded lower abundance than net and pump sampling. There was no significant difference in the taxa richness yielded from the three methods or in the total abundance from net and pump samples.

The macroinvertebrate communities found at deep sites (>4 m) were distinct from those recorded in shallower areas. These deep sites had lower species richness and abundance, and were dominated by low-oxygen tolerant taxa. The presence of such communities in deeper areas should be considered when examining biotic assemblages for evidence of nutrient enrichment within Lough Carra. No distinction was found between invertebrate assemblages in shallow (<2 m) and mid-depth sites (2 -4 m).

Aside from depth, no significant relationship was found between any of the abiotic variables measured and the invertebrate distribution. The nutrient concentration did not vary greatly across sites and the biotic community present is known to have a broad tolerance to nutrient concentration. This may account for the absence of a significant relationship between nutrients and invertebrate distribution. Species richness and in particular total abundance of macroinvertebrates did vary greatly among sites. Therefore, it is likely that abiotic factors are influencing distribution. More specific research, analysing individual environmental factors, may be needed to identify the relationship between abiotic pressure and invertebrate community response, within the lake.

Very few pollution-sensitive species were recorded across all samples sites. Even when the low number of mesohabitats surveyed is considered, the macroinvertebrate community composition recorded is not reflective of a pristine lake.

A significant relationship exists between water depth and sediment chemistry in Lough Carra. The highest values for Fe, Mg, TP, TN and TOC were all recorded in deep water sites. A lower accumulation rate of $CaCO_3$ in deep water sediment is the likely explanation for this. The relationship between sediment chemistry and depth must be considered when comparing chemical values both within and among lakes. The influence of depth is particularly important when examining variation in nutrient content among the three lake basins.

A positive relationship exists between total phosphorus and iron and manganese within the sediment, most likely owing to binding of TP to these elements. The Fe:TP ratio in the sediment is far lower than was recorded in 2002. This suggests that the capacity of the lake sediment to absorb nutrients has been reduced in recent years. Further analysis concerning the nutrient binding in the sediment is necessary to confirm if this is the case. If nutrient input to the lake continues at the current rate, the potential exits for complete P saturation of the sediment and subsequent rapid decline in water quality. This could set in motion a series of natural feed-back mechanisms which would severely hamper any attempts at remediation.

## References


BACHMANN, V. & USSEGLIO-POLATERA, P. (1999) Contribution of the macrobenthic compartment to the oxygen budget of a large regulated river: the Mosel. *Hydrobiologia,* 410**,** 39-46.

BAUMGARTNER, D., MORTL, M. & ROTHHAUPT, K. O. (2008) Effects of water-depth and water-level fluctuations on the macroinvertebrate community structure in the littoral zone of Lake Constance. *Hydrobiologia,* 613**,** 97-107.

BRAUNS, M., GARCIA, X. F., PUSCH, M. T. & WALZ, N. (2007) Eulittoral macroinvertebrate communities of lowland lakes: discrimination among trophic states. *Freshwater Biology,* 52**,** 1022-1032.

BRONMARK, C. & HANSSON, L.-A. (2005) The Biology of Lakes and Ponds*,* New York, Oxford University Press.

CASCORBI, U. (2002) The influence of land use systems on substrate patterns in brooks and their benthic macroinvertebrate microdistribution. *Geoderma,* 105**,** 179-200.

CHAVES, M. L., CHAINHO, P. M., COSTA, J. L., PRAT, N. & COSTA, M. J. (2005) Regional and local environmental factors structuring undisturbed benthic macroinvertebrate communities in the Mondego River basin, Portugal. *Archiv Fur Hydrobiologie,* 163**,** 497-523.

CLARKE, K. R. & WARWICK, R. M. (2001) *Change in marine communities: an approach to statistical analysis and interpretation,* Plymouth, PRIMER-E.

CORKUM, L. D., CIBOROWSKI, J. J. H. & DOLAN, D. M. (2006) Timing of Hexagenia (Ephemeridae : Ephemeroptera) mayfly swarms. *Canadian Journal of Zoology-Revue Canadienne De Zoologie,* 84**,** 1616-1622.

COSTELLO, M. J. (1993) Biogeography of Alien Amphipods Occurring in Ireland, and Interactions with Native Species. *Crustaceana,* 65**,** 287-299.

CZACHOROWSKI, S. (1993) Distribution of Trichoptera larvae in vertical profile of lakes *Polskie Archiwum Hydrobiologii,* 40**,** 139-163.

DERMOTT, R. (1978) Benthic Diversity and Substrate-Fauna Associations in Lake Superior. *Journal of Great Lakes Research,* 4**,** 505-512.

DICK, J. T. A. (1996) Post-invasion amphipod communities of Lough Neagh, Northern Ireland: Influences of habitat selection and mutual predation. *Journal of Animal Ecology,* 65**,** 756-767.

DITTRICH, M. & KOSCHEL, R. (2002) Interactions between calcite precipitation (natural and artificial) and phosphorus cycle in the hardwater lake. *Hydrobiologia,* 469**,** 49-57.

DŽEROSKI, S., DEMŠAR, D. & GRBOVIĆ, J. (2000) Predicting Chemical Parameters of River Water Quality from Bioindicator Data. *Applied Intelligence,* 13**,** 7-17.

ELEXOVÁ, E. & NÉMETHOVÁ, D. (2003) The effect of abiotic environmental variables on the Danube macrozoobenthic communities. *Limnologica - Ecology and Management of Inland Waters,* 33**,** 340-354.

ELLIOT, J. M., HUMPESCH, U. H. & MACAN, T. T. (1988) *Larvae of the British Ephemeroptera: a key with ecological notes.*, Freshwater Biological Association Scientific Publication No. 49.

ELLIOT, J. M. & MANN, K. H. (1979) *A key to the British freshwater leeches with notes on their life cycles and ecology.*, Freshwater Bioogical Association Scientific Publication No. 40.

ELLISON, M. E. & BRETT, M. T. (2006) Particulate phosphorus bioavailability as a function of stream flow and land cover. *Water Research,* 40**,** 1258-1268.

ENGSTROM, D. R. & WRIGHT, H. E. (1984) *Chemical stratigraphy of lake sediments as a record of environmental change,* Leicester, Leicester University Press.

FITTER, R. & MANUEL, R. (1994) *Collins Photo Guide: Lakes, Rivers, Streams and Ponds of Britain and North-West Europe,* London, HarperCollins.

FREE, G., LITTLE, R., TIERNEY, D., DONNELLY, K. & CARONI, R. (2006) A reference based typology and ecological assessment system for Irish lakes. Environmental Protection Agency, Ireland.

FRIDAY, L. E. (1988) *A key to the British Water Beetles.* , Field Studies 7.

FULLER, R. L., KENNEDY, B. P. & NIELSEN, C. (2004) Macroinvertebrate responses to algal and bacterial manipulations in streams. *Hydrobiologia,* 523**,** 113-126.

GLEDHILL, T., SUTCLIFFE, D. W. & WILLIAMS, W. D. (1993) *British Freshwater Crustacea Malacostraca. A key with ecological notes.* Freshwater Biological Association Scientific Publication No. 52.

GONZALEZ, J. M., BASAGUREN, A. & POZO, J. (2001) Life history and production of Caenis luctuosa (Burmeister) (Ephemeroptera, Caenidae) in two nearby reaches along a small stream. *Hydrobiologia,* 452**,** 209-215.

GRACA, M. A. S., PINTO, P., CORTES, R., COIMBRA, N., OLIVEIRA, S., MORAIS, M., CARVALHO, M. J. & MALO, J. (2004) Factors affecting macroinvertebrate richness and diversity in Portuguese streams: a two-scale analysis. *International Review of Hydrobiology,* 89**,** 151-164.

GRAY, N. F. (2005) Water Technology. An introduction for environmental scientists and engineers, Oxford, Elsevier.

GRIFFIOEN, J. (2006) Extent of immobilisation of phosphate during aeration of nutrient-rich, anoxic groundwater. *Journal of Hydrology,* 320**,** 359-369.

GUNNARS, A., BLOMQVIST, S., JOHANSSON, P. & ANDERSSON, C. (2002) Formation of Fe(III) oxyhydroxide colloids in freshwater and brackish seawater, with incorporation of phosphate and calcium. *Geochimica et Cosmochimica Acta,* 66**,** 745-758.

HARKER, J. E. (1992) SWARM BEHAVIOR AND MATE COMPETITION IN MAYFLIES (EPHEMEROPTERA). *Journal of Zoology,* 228**,** 571-587.

HEATH, C. R., LEADBEATER, B. C. S. & CALLOW, M. E. (1995) EFFECT OF INHIBITORS ON CALCIUM-CARBONATE DEPOSITION MEDIATED BY FRESH-WATER ALGAE. *Journal of Applied Phycology,* 7**,** 367-380.

HEINO, J. (2000) Lentic macroinvertebrate assemblage structure along gradients in spatial heterogeneity, habitat size and water chemistry. *Hydrobiologia,* 418**,** 229-242.

HOBBS, W., IRVINE, K. & DONOHUE, I. (2005) Using sediments to assess the resistance of a calcareous lake to diffuse nutrient loading. *Archiv Fur Hydrobiologie,* 164**,** 109-125.

HUMPHRIES, P., GROWNS, J. E., SERAFINI, L. G., HAWKING, J. H., CHICK, A. J. & LAKE, P. S. (1998) Macroinvertebrate sampling methods for lowland Australian rivers. *Hydrobiologia,* 364**,** 209-218.

HUXLEY, C. & THORTON, J. (2004) Landuse Changes in the Lough Carra Catchment. IN HUXLEY, C. & IRVINE, K. (Eds.) *The Great Western Lakes, Ecology, Heritage and Management* Castlebar, Galway and Mayo Insitute of Technology, Trinity College.

IRVINE, K., DONOHUE. I, WEMAERE. A, D, S. & HOBBS, W. (2003) CONNECTING LAND-USE WITH WATER QUALITY: SOURCES, SINKS AND TIMEBOMBS. *International Water Association, Diffuse Pollution Conference Proceedings, Dublin*.

JAMES, M. R., HAWES, I., WEATHERHEAD, M., STANGER, C. & GIBBS, M. (2000) Carbon flow in the littoral food web of an oligotrophic lake. *Hydrobiologia,* 441**,** 93-106.

JIN, X., WANG, S., PANG, Y. & CHANG WU, F. (2006) Phosphorus fractions and the effect of pH on the phosphorus release of the sediments from different trophic areas in Taihu Lake, China. *Environmental Pollution,* 139**,** 288-295.

JORDAN, P. & RIPPEY, B. (2003) Lake sedimentary evidence of phosphorus, iron and manganese mobilisation from intensively fertilised soils. *Water Research,* 37**,** 1426-1432.

KALLER, M. D., HARTMAN, K. J. & ANGRADI, T. R. (2001) Experimental determination of benthic macro invertebrate metric sensitivity to fine sediment in Appalachian streams. IN EVERSOLE, A. G. (Ed. *55th Annual Conference of the Southeastern-Association-of-Fish-and-Wildlife-Agencies.* Louisville, Ky.

KATSEV, S., TSANDEV, I., L'HEUREUX, I. & RANCOURT, D. G. (2006) Factors controlling long-term phosphorus efflux from lake sediments: Exploratory reactive-transport modeling. *Chemical Geology,* 234**,** 127-147.

KELLY-QUINN, M. & BRACKEN, J. J. (2000) The distribution of the Ephemeroptera in Ireland. . *Occasional Publication of the Irish Biogeographical Society,* 5**,** 223.

KING, J. J. & CHAMP, W. S. T. (2000) Baseline water quality investigations on Lough Carra, western Ireland, with reference to water chemistry, phytoplankton and aquatic plants. *Biology and Environment-Proceedings of the Royal Irish Academy,* 100B**,** 13-25.

KOLAR, C. S. & RAHEL, F. J. (1993) Interaction of a biotic factor (predator presence) and an abiotic factor (low-oxygen) as an influence on benthic invertebrate communities. *Oecologia,* 95**,** 210-219.

KRANTZBERG, G. (1985) The influence of bioturbation on physical, chemical and biological parameters in aquatic environments: A review. *Environmental Pollution Series A, Ecological and Biological,* 39**,** 99-122.

LANDA, V. & SOLDAN, T. (1985) Distributional patterns, chorology and origin of the czechoslovak fauna of mayflies (ephemeroptera). *Acta Entomologica Bohemoslovaca,* 82**,** 241-268.

LÖFGREN, S. & BOSTRÖM, B. (1989) Interstitial water concentrations of phosphorus, iron and manganese in a shallow, eutrophic swedish lake--Implications for phosphorus cycling. *Water Research,* 23**,** 1115-1125.

MACAN, T. T. (1977) *A key to the British fresh and brackish water Gastropods with notes on their ecology.*, Freshwater Biological Association Scientific Publication No. 13.

MACNEIL, C., BIGSBY, E., DICK, J. T. A., HYNES, H. B. N., HATCHER, M. J. & DUNN, A. M. (2003a) Temporal changes in the distribution of native and introduced freshwater amphipods in Lough Neagh, Northern Ireland. *Archiv Fur Hydrobiologie,* 157**,** 379-395.

MACNEIL, C., DICK, J. T. A. & ELWOOD, R. W. (2000) Differential physico-chemical tolerances of amphipod species revealed by field transplantations. *Oecologia,* 124**,** 1-7.

MACNEIL, C., DICK, J. T. A., HATCHER, M. J., TERRY, R. S., SMITH, J. E. & DUNN, A. M. (2003b) Parasite-mediated predation between native and invasive amphipods. *Proceedings of the Royal Society of London Series B-Biological Sciences,* 270**,** 1309-1314.

MATISOFF, G. & NEESON, T. M. (2005) Oxygen Concentration and Demand in Lake Erie Sediments. *Journal of Great Lakes Research,* 31**,** 284-295.

MCCARTHY, T., K, R. BARBIERO. R, M. O'CONNELL, M. GUIRY, D. DOHERTY, P. CULLEN, J. KING, B. O'CONNOR. (1997) Investigation of Eutrophication Processes in the Littoral Zones ofWestern Irish Lakes. *Environmental Protection Agency, Ardcavan, Wexford, Ireland*.

MCGOFF, E. & IRVINE, K. (2009) A test of the association between Lake Habitat Quality Assessment and macroinvertebrate community structure. *Aquatic Conservation-Marine and Freshwater Ecosystems,* 19**,** 520-533.

MEIJER, L. E. & AVNIMELECH, Y. (1999) On the use of micro-electrodes in fish pond sediments. *Aquacultural Engineering,* 21**,** 71-83.

MIYAKE, Y. A. S. N. (2002) Effects of substratum stability on diversity of stream invertebrates during baseflow at two spatial scales. *Freshwater Biology,* 47**,** 219-230.

MOSS, B. (2000) *Ecology of Freshwaters. Man and Medium, Past to Future,* Oxford, Blackwell Science Ltd.

MULLER, B., WANG, Y., DITTRICH, M. & WEHRLI, B. (2003) Influence of organic carbon decomposition on calcite dissolution in surficial sediments of a freshwater lake. *Water Research,* 37**,** 4524-4532.

O'TOOLE, C., DONOHUE, I., MOE, S. J. & IRVINE, K. (2007) Nutrient optima and tolerances of benthic invertebrates, the effects of taxonomic resolution and testing of selected metrics in lakes using an extensive European data base. *Conference on Relationships Between Ecological and Chemical Status of Surface Waters (REBECCA).* Oslo, NORWAY, Springer.

RAUNIO, J., IHAKSI, T., HAAPALA, A. & MUOTKA, T. (2007) Within- and among-lake variation in benthic macroinvertebrate communities - comparison of profundal grab sampling and the chironomid pupal exuvial technique. *Journal of the North American Benthological Society,* 26**,** 708-718.

RIERADEVALL, M., BONADA, N. & PRAT, N. (1999) Substrate and depth preferences of macroinvertebrates along a transect in a Pyrenean high mountain lake (Lake Redó, NE Spain). *Limnetica* 17**,** 127-134.

ROSENBERG, D. M. & RESH, V. H. (1993) *Freshwater Biomonitoring and Benthic Macroinvertebrates,* London, Chapman and Hall.

RUBBO, M. J., BELDEN, L. K. & KIESECKER, J. M. (2008) Differential responses of aquatic consumers to variations in leaf-litter inputs. *Hydrobiologia,* 605**,** 37-44.

SCHLOESSER, D. W. & NALEPA, T. F. (2002) Comparison of 5 benthic samplers to collect burrowing mayfly nymphs (Hexagenia spp : Ephemeroptera : Ephemeridae) in sediments of the Laurentian Great Lakes. *Journal of the North American Benthological Society,* 21**,** 487-501.

SMITH, R. V., JORDAN, C. & ANNETT, J. A. (2005) A phosphorus budget for Northern Ireland: inputs to inland and coastal waters. *Journal of Hydrology,* 304**,** 193-202.

STOFFELS, R. J., CLARKE, K., R & CLOSS. G, P. (2005) Spatial scale and benthic community organisation in the littoral zones of large oligotrophic lakes: potential for cross-scale interactions. *Freshwater Biology,* 50**,** 1131-1145.

SYROVATKA, V., SCHENKOVA, J. & BRABEC, K. (2009) The distribution of chironomid larvae and oligochaetes within a stony-bottomed river stretch: the role of substrate and hydraulic characteristics. *Fundamental and Applied Limnology,* 174**,** 43-62.

TAKAMURA, N., ITO, T., UENO, R., OHTAKA, A., WAKANA, I., NAKAGAWA, M., UENO, Y. & NAKAJIMA, H. (2009) Environmental gradients determining the distribution of benthic macroinvertebrates in Lake Takkobu, Kushiro wetland, northern Japan. *Ecological Research,* 24**,** 371-381.

TAYLOR, D., JORDAN, P., LEIRA, M. & DALTON, C. (2007) Identification of reference - status of Irish lake typologies using paeleolimnological methods and techniques (IN-SIGHT). Johnstown Castle, Co. Wexford, Environmental Protection Agency.

TOLONEN, K. T., HAMALAINEN, H., HOLOPAINEN, I. J. & KARJALAINEN, J. (2001) Influences of habitat type and environmental variables on littoral macroinvertebrate communities in a large lake system. *Archiv Fur Hydrobiologie,* 152**,** 39-67.

TRIGAL, C., GARCIA-CRIADO, F. & ALAEZ, C. F. (2007) Macroinvertebrate communities of mediterranean ponds (North Iberian Plateau): importance of natural and human-induced variability. *Freshwater Biology,* 52**,** 2042-2055.

TRIGAL, C., GARCIA-CRIADO, F. & FERNANDEZ-ALAEZ, C. (2006) Among-habitat and temporal variability of selected macroinvertebrate based metrics in a Mediterranean shallow lake (NW spain). *Hydrobiologia,* 563**,** 371-384.

VIRGINIA, S.-E., MA ANGELES GARCÍA DEL, C., CARMEN, A., ASUNCIÓN DE LOS, R., JACEK, W. & JUAN ANTONIO, G.-M. (2006) Interaction between Calcite and Phosphorus in Biomineralization Processes in Tufa Carbonates. *International Review of Hydrobiology,* 91**,** 222-241.

WALLACE, I. D., WALLACE, B. & PHILIPSON, G. N. (1990) *A key to the case-bearing caddis larvae of Britain and Ireland.*, Freshwater Biological Association Scientific Publication No. 51.

WALLACE, J. B., EGGERT, S. L., MEYER, J. L. & WEBSTER, J. R. (1999) Effects of resource limitation on a detrital-based ecosystem. *Ecological Monographs,* 69**,** 409-442.

WHITE, J. & IRVINE, K. (2003) The use of littoral mesohabitats and their macroinvertebrate assemblages in the ecological assessment of lakes. *Aquatic Conservation: Marine and Freshwater Ecosystems,* 13**,** 331-351.

WOHL, D. L., WALLACE, J. B. & MEYER, J. L. (1995) Benthic macroinvertebrate community structure, function and production with respect to habitat type, reach and drainage basin in the southern Appalachians (USA). *Freshwater Biology,* 34**,** 447-464.

WWW.EPA.IE. www.epa.ie/downloads/pubs/water/waterqua/EPA_water_quality_2001-3.pdf

WWW.NPWS.IE. www.npws.ie/en/media/Media,4107,en.pdf

YO MIYAKE, S. N. (2002) Effects of substratum stability on diversity of stream invertebrates during baseflow at two spatial scales. *Freshwater Biology,* 47**,** 219-230.