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THE NUTRIENT ASSIMILATIVE CAPACITY OF MAERL AS A SUBSTRATE IN CONSTRUCTED WETLAND SYSTEMS FOR WASTE TREATMENT

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Abstract—This study evaluated the performance of maerl (calcified seaweed) as a substrate for artificial wetland waste treatment systems. Pilot scale artificial wetlands were set up in the laboratory; three planted with *Phragmites australis* and three unplanted, and fed with a synthetic sewage solution. The effluent from the tanks was monitored over a period of 9 weeks for chemical oxygen demand, total nitrogen, total phosphorus, ammonium-N, total oxidised nitrogen, soluble reactive phosphorus, dissolved oxygen, pH and temperature. The data were analysed using repeated measures ANOVA to look for significant differences between treatments, and within treatments, over time. A batch incubation experiment was also carried out to ascertain the maximum adsorption capacity of maerl for phosphorus. Results obtained were compared with those in the literature for other substrates. Variability within and between treatments was high, but it was found that maerl effectively removed total phosphorus (98%). Nitrogen removal was less effective, with the tanks producing ammonium-N. The low nitrogen removal shown in the tanks was a factor of the short duration of the experiment; but ammonification did decrease in the planted tanks over time. Performance at removing nitrogen was normal when compared with figures in the literature, but phosphorus removal by maerl was considerably higher than gravel bed wetlands, and comparable with the very best figures given for artificial wetlands based on novel substrates such as shales and slags. This trial showed that maerl has great potential as a constructed wetland substrate, due to its high phosphorus-adsorbing capacity.

Key words—constructed wetlands, maerl, wastewater, phosphorus, nitrogen

INTRODUCTION

Natural wetlands have been used for centuries as a sink for waste, being capable of assimilating large amounts of environmental contaminants (Dinges, 1982). Constructed wetland systems seek to emulate the properties of natural wetlands in an environment which can be controlled and manipulated. These systems rely on several basic processes to purify contaminated wastewater: uptake of nutrients by plants; bacterial degradation and oxidation of contaminants; sedimentation; and adsorption of particles and dissolved substances in the waste on to the substrate (Reed *et al.*, 1988).

The ability to remove pollutants varies widely within and between constructed wetland systems, but their general performance can compare favourably with conventional treatment systems. Removal of biochemical oxygen demand (BOD), chemical oxygen demand (COD), faecal coliforms and suspended solids for example, can approach 100% in artificial wetlands, (Tanner *et al.*, 1995; von Felde and Kunst, 1997), although removal of nitrogen and phosphorus has tended to be variable and often unsatisfactorily low (Wood, 1995).

Nitrogen removal chiefly occurs within the rhizosphere of the wetland, where large populations of sessile anaerobic and aerobic bacteria grow, adsorbing and breaking down the organic and inorganic components of sewage (Flemming, 1995). Total nitrogen ranges from 20 to 70 mg/l in domestic sewage (Horan, 1990), and is present in four forms: organic nitrogen; nitrite (NO_2-N); nitrate (NO_3-N); and ammonium (NH_4-N). Organic nitrogen, comprising a variety of compounds such as amino acids and urea (Kadlec and Knight, 1996), and NH_4-N are found in the greatest concentrations, the latter being found at about 20 mg/l in sewage (Horan, 1990).

NH_4-N, itself largely derived from organic N

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through the process of ammonification, is oxidised to NO_2-N and NO_3-N (known collectively as total oxidised nitrogen, TON) by nitrifying bacteria which utilise the aerobic conditions found around the roots of wetland plants (Brix, 1997). Nitrification can be slow, and in some cases fails completely, with levels of NH_4-N actually increasing within the wetland as a result of high organic N concentrations in the influent being aerobically degraded without subsequent oxidation (Reed and Brown, 1995).

TON diffuses away from the aerobic rhizosphere into the anaerobic water-saturated zone of the wetland and is then permanently lost from the system either through denitrification to nitrogen gas, which is released to the atmosphere (McEldowney *et al.*, 1993), or through leaching. Removal of total nitrogen in constructed wetlands is often low, 46–72% on average (Hammer and Knight, 1994), and in many cases is not sufficient to meet discharge consents (see for example Boutin *et al.*, 1997). Generally poor removal of nitrogen is attributed to nitrification being limited by low oxygen and high carbon concentrations derived from the influent sewage.

Although the biota are responsible for removing a large proportion of most contaminants, they are not capable of removing significant amounts of phosphorus. Phosphorus is present in sewage at levels of between 6 and 10 mg/l, while less than 10 mg/l in the environment may stimulate algal growth (Horan, 1990). It has been found however, that the choice of substrate is crucial for maximising phosphorus removal from waste water, as abiotic interactions between P and sediment particles, including the processes of sedimentation and adsorption, are the major mechanisms for the removal of P.

The P-adsorption capacity of a wetland is dependent on two factors: the number of adsorption sites, proportional to the surface area of substrate particles and pH. Phosphorus adsorption is highest in alkaline wetlands, containing large amounts of calcium, and acid wetlands, containing large concentrations of aluminium and iron (McEldowney *et al.*, 1993), phosphorus being precipitated by reactions with these elements. For example, calcium reacts with soluble phosphorus at high pH to form hydroxyapatite, which precipitates out of solution (Stumm and Morgan, 1981).

Different wetland substrates have different characteristics, and thus different assimilative properties, phosphorus adsorption capacity being determined by a compromise between hydraulic conductivity and surface area. A large hydraulic conductivity, necessary to avoid clogging, often precludes a large substrate surface area, resulting in low phosphorus adsorption capability. Natural soils for example, commonly used as wetland substrates, can remove up to 98% of influent phosphorus (Dinges, 1982), but tend to clog due to low hydraulic conductivity (Cooper, 1993).

River gravels are also commonly used as constructed wetland substrates. Gravels have a high hydraulic conductivity, but their capacity for phosphorus removal is generally low; the particles are impermeable, and have a low surface area compared to volume. Figures quoted for the removal of phosphorus by gravel wetlands range from as low as 20% (Fisher, 1988) to over 90% with certain types of iron rich gravel (Reed and Brown, 1995).

There is a current trend towards the use of novel products as constructed wetland substrates, which has resulted in several studies to investigate the efficiency of industrial by-product substrates for the removal of phosphorus (see for example, Drizo *et al.*, 1997; Lopez *et al.*, 1998).

The study described here proposes the use of maerl as a potential constructed wetland substrate. Maerl is the dead deposits of calcareous red algae (Corallinaceae) found growing in shallow waters around the coast of north-west Europe and the western Mediterranean. Upon death the algae accumulate to form large shoals, providing an important habitat for benthic communities (Hall-Spencer, 1998).

The use of maerl exploits its high mineral content; as well as high percentages of calcium carbonate (80%) and magnesium carbonate (10%), it contains small amounts of 25 different trace elements such as sulphur (0.6%), phosphorus (0.35%), potassium (0.2%), sodium (0.17%), iron (2500 ppm), aluminium (500 ppm) and copper (15 ppm) (Inglethorpe, 1992).

It was hypothesised that maerl would be an effective constructed wetland substrate for removing phosphorus from effluent for three main reasons: it has a large surface area to volume ratio of 2200 m^2 l^{-1} (Inglethorpe, 1992); it is highly porous; and it has a high calcium carbonate content. A laboratory experiment was conducted in order to investigate the validity of this hypothesis. The experiment involved monitoring pilot scale artificial wetlands set up in the laboratory, for several parameters over a period of several weeks. The specific aims were: to investigate the suitability of maerl as a wetland substrate to support growth of the common reed, *Phragmites australis*; to investigate the ability of maerl to purify waste water; to determine the equilibrium phosphorus absorption capacity of maerl, in order to estimate the lifetime of an actual constructed wetland system in the field, and; to compare the performance of maerl as a substrate with published data on the performance of other substrates.

MATERIALS AND METHODS

Phosphorus-adsorption capacity of maerl

The P-adsorption capacity of maerl was determined by

Table 1. Summary of the variables measured in the study

Variable	Method of determination	Sample storage	Reference
pH	pH meter	Not stored	
Dissolved oxygen	Dissolved oxygen meter (YSI 59)	Not stored	
Chemical oxygen demand	HACH reactor digestion, then spectrophotometer	Not stored	Anon (1993)
Total phosphorus	Persulphate digestion, then ammonium molybdate method	In fridge (24 h)	APHA (1995)
Soluble reactive phosphorus	Ammonium molybdate method	Frozen	APHA (1995)
Total nitrogen	Persulphate digestion, then hydrazine reduction method	In fridge (24 h)	APHA (1995) (modification)
Ammonium-N	Alkaline phenol hypochlorite method	Frozen	Mackereth *et al.* (1978)
Total oxidised nitrogen	Hydrazine reduction	Frozen	APHA (1995) (modification)

a batch incubation experiment, designed to compare the performance of maerl with that of river gravel, by equilibrating a small quantity of substrate within a range of orthophosphate-P solutions (made from KH_2PO_4), a method adapted from those used by Zhu *et al.* (1997) and Sakadevan and Bavor (1998). Conical flasks containing 4 g of each sample, plus a set of flasks with no substrate (blanks) were set up in triplicate, each with 100 ml of P solution (0, 5, 50, 500 and 5000 mg l^{-1}). Two drops of chloroform, to inhibit microbial growth, were added to each flask, which were then shaken on a mechanical shaker at 200 rpm for 72 h. A 20 ml sample was taken from each flask at 24, 48 and 72 h. These were centrifuged at 4000 rpm for 10 min, and the supernatant analysed for soluble reactive phosphorus (SRP) by the stannous chloride method (APHA 1995).

The P retained by the substrate was converted into P adsorption in mg kg^{-1} using equation (1) as given by Reddy *et al.* (1998), and used to estimate the lifetime of a maerl constructed wetland system:

$$\text{P adsorption}\,(\text{mg kg}^{-1}) = \frac{[(C_o \times V) - (C_t \times V)]}{M} \qquad (1)$$

where C_o is concentration of P (mg l^{-1}) added, V is volume of liquid (l), C_t is concentration of P in solution after time period (mg l^{-1}), and M is mass of dry substrate used (kg).

MAERL AS A CONSTRUCTED WETLAND SUBSTRATE

Experimental design

Six tanks, each with a surface area of 1.29 m^2, were filled with approximately 21 l volume (approximately 21 kg) of fine grade rinsed maerl and placed under five fluorescent lights on a photoperiod of 12 h light–12 h dark. The two treatments—three unplanted and three planted, each with ten seedlings of the Common Reed (*P. australis*)—were allocated randomly to the six tanks.

A synthetic sewage solution was used as the influent for the tanks to minimise variables within the experiment (OECD standard synthetic sewage, as given by Drizo *et al.* (1997)). The sewage concentrate was diluted 100-fold and added to a 65 l reservoir (thermostatically controlled to 0–4°C) every 2 days. On average, 21 l of the maerl held 11.5 l of water (pore volume of 55%); therefore a flow rate of 2.3 l per tank per day was needed to achieve a hydraulic retention time (HRT) of 5 days. This was controlled using a multichannel peristaltic pump.

At the start of the experiment, each tank was filled with 11.5 l of the dilute sewage solution, and sewage inflow started immediately. Each tank was inoculated with 14 ml of faecal microorganism-free activated sludge effluent, from a model activated sludge plant. First sampling of the effluent from the tanks took place 5 days after inoculation, and subsequently at seven day intervals, samples being taken in dedicated 200 ml acid-washed plastic bottles, and analysed without filtration. Concentrated sewage and diluted sewage (1:100) from the reservoir were also analysed.

Analyses

A total of eight variables were measured in the sewage and tank effluent samples collected during the experiment (Table 1). These parameters aimed to provide a data set which comprehensively characterised the quality of the effluent, and hence the performance of the tanks.

Data analysis

This experiment was set up as a mixed factorial experiment, one treatment factor being between subject tanks (planted versus unplanted), and one factor within subject tanks (time), thus compatible with analysis using Repeated Measures ANOVA within the statistical programme SPSS (Kinnear and Gray, 1997). This type of ANOVA is appropriate in any experiment where data derived from a normal population are taken on the same subjects repeatedly over a period of time (Manly, 1992). Data of this type are not independent, therefore ordinary t-tests and ordinary ANOVA are not suitable. Results were taken to be significant at the 5% level ($P = 0.05$). The null hypotheses tested by the analysis were: planted and unplanted tanks showed no differences in performance; and that there was no change in performance within treatments over time.

RESULTS AND DISCUSSION

Phosphorus-adsorption capacity of maerl

Equilibrium was reached in the flasks of maerl after 48 h, and for 5 and 50 mg l^{-1} P solutions, P was almost completely removed (Table 2). Removal was less efficient at higher concentrations. The gravel samples removed no P from the solutions

Table 2. Summary of results of batch incubation experiment after 48 h

Initial P concentration (mg l^{-1})	Final P concentration (mg l^{-1})	% Removal	Removal (mg kg)
0	0	0	0
5	0.1	99	123
50	2.6	95	1184
500	200	60	7490
5000	3420	32	39,500

Table 3. Summary of experimental results (all values in mg l^{-1} unless otherwise stated)

Parameter	Dilute sewage	Standard deviation	Planted tanks	Standard deviation	Unplanted tanks	Standard deviation
DO	11.8	0.2	1.2[a]	0.2	1.5[a]	0.4
pH (units)	6.8		7.6		7.6	
COD	331	21	85[a]	48	174[a]	52
TN	47.2	5.3	19.4[a]	8	28.2[a]	9
NH_4-N	3.2	0.4	24.5[b]	6	30.9[a]	3
TON	0.10	0.02	0.09	0.03	0.08	0.09
TP	7.5	1.2	0.14[a]	0.07	0.19[a]	0.09
SRP	6.96	1.3	0.05[a]	0.02	0.06[a]	0.04

[a]Indicates a significant difference from concentration in influent sewage.
[b]Indicates a significant difference between planted and unplanted tanks.

and were therefore disregarded in subsequent analysis. Since the maerl removed over 95% of the phosphorus from the 50 mg l^{-1} solution, the equilibrium adsorption capacity was estimated at 1184 mg P kg^{-1} using equation (1).

This value can be used to estimate the length of time before a maerl constructed wetland would become saturated with phosphorus. In each tank in this experiment there was approximately 21 kg of maerl receiving 113 mg P per week. Assuming this was totally removed from the effluent, 5.4 mg P kg^{-1} maerl was adsorbed over a week. The equilibrium value for maerl was 1184 mg kg^{-1}, therefore the substrate would be saturated after 219 weeks (4 years) of sewage input with a phosphorus concentration of 7.5 mg l^{-1}. This estimate does not take account of plant uptake and long-term sedimentation and burial processes, and therefore the length of time to substrate saturation may actually be longer.

The adsorption capacity of maerl for phosphorus compares favourably with those quoted in the literature for other substrates. Gravel has a low P-adsorbing capacity, less than 50 mg kg^{-1} (Breen, 1990), while some of the industrial by-products have an extremely high P-adsorbing capacity e.g. the blast furnace slag tested by Sakadevan and Bavor (1998), had a P-adsorbing capacity of over 44,000 mg kg^{-1}. Lightweight Expanded Clay Aggregates (LECA) tested by Reddy *et al.*, (1998) were found to have variable P adsorbing properties depending on the soil of origin, ranging from 37 to 2900 mg.kg^{-1}.

Sakadevan and Bavor (1998), estimated that a constructed wetland built using industrial slags as a substrate would have a lifetime of at least 3.5 years, comparable with the performance of maerl. In fact, the amount of P applied to the system during the 9 weeks of study (1017 mg P per tank) was only 4% of the theoretical adsorption maximum obtained from the batch incubation experiment. A study involving increasing the concentrations of P in the synthetic sewage solution, in order to compare saturation times, would help to confirm these conclusions.

REMOVAL OF NUTRIENTS FROM SEWAGE BY TANKS

Reed growth was sustained throughout the experimental period, with evidence of horizontal spreading of rhizomes, although all plants became infested by aphids.

All tanks were quickly colonised by algae, of various species and growth form, including blue-greens, filamentous green algae and diatoms. In the planted tanks, however, algal cover was limited by the grazing actions of snails imported inadvertently with the reed plants. Two species of snail were present; the great pond snail (*Lymnaea stagnalis*) and the wandering snail (*L. peregra*), and both populations quickly increased, with many clusters of eggs visible at the base of each reed plant. Uptake of nutrients by the snails (from the algae) and the aphids (from the reeds) must have been considerable, due to the high numbers present.

The mean concentration of contaminants in the sewage and the effluent from the tanks are summarised in Table 3. It can be seen that five out of eight parameters measured were reduced in concentration by both planted and unplanted tanks. pH increased, as did ammonium-N concentration. TON concentration was not changed significantly within the tanks from amounts found in the influent sewage. The differences between planted and unplanted tanks were not great, with only NH_4-N showing significant differences between treatments. P was almost completely removed by both treatments. The average temperature of the tanks was 21°C.

Removal of chemical oxygen demand

Although planted tanks removed more COD on average from the sewage than unplanted tanks (75% removal compared to 48%), this difference was not significant (P = 0.07), due to the high variability of the data. However, the pattern of COD removal over time did change significantly (P = 0.009). Figure 1 shows that planted tanks improved consistently in COD removal over the duration of the experiment, whilst the unplanted tanks showed a less consistent trend.

Nitrogen removal

The sewage solution contained 47.2 mg TN l^{-1}, 6.8% of which was accounted for by NH_4-N, (3.2 mg l^{-1}), 0.2% by TON, leaving 93% in the form of particulate or soluble organic N. This low NH_4-N concentration is unlike that found in real sewage.

On average, planted tanks removed 59% of the incoming nitrogen, while unplanted tanks removed less, at 41%. The performance of the two treatments was not significantly different (between groups $P = 0.13$, over time $P = 0.16$).

Concentrations of TON in the incoming sewage were low, 97 μg l^{-1}. Concentrations in the tank samples were similar to those in the influent sewage, and there were no significant differences between the two treatments ($P = 0.3$), or over time ($P = 0.34$).

The total nitrogen present in the sewage solution as organic N was largely converted to NH_4-N within the tanks. NH_4-N concentrations in the tank samples were 5–10 times higher than those found in the sewage solution, showing that significant ammonification was occurring. Although planted and unplanted tanks showed similar results at the beginning of the experiment (Fig. 2), with successive weeks the planted tanks showed significant differences in performance ($P = 0.005$), producing progressively less NH_4-N, with a mean concentration of 24.5 mg l^{-1} over the entire time period. The unplanted tanks showed less of a decrease in NH_4-N production over time with a significantly higher mean concentration of 30.9 mg l^{-1} ($P = 0.03$). For this parameter, the null hypothesis was rejected at the 95% confidence level.

The decreases observed in the production of NH_4-N over time were in spite of the production of NH_4-N waste products by the flora and fauna present. The reduction in NH_4-N over time may have been brought about by a combination of three factors:

1. nitrification of NH_4-N to NO_3-N and NO_2-N at aerobic plant roots, with subsequent rapid denitrification to the atmosphere in the anaerobic parts of the substrate;
2. increasing uptake of NH_4-N by the biota;
3. increasing sedimentation of particulate organic N enabled by the development of root and microbial biomass.

There was little evidence of significant nitrification. Firstly, observed TON concentrations in the tank effluent were similar to those of the influent sewage. Secondly, dissolved oxygen levels in the effluent were very low (less than 2 mg l^{-1}), suggesting that the reeds were not producing the significant amounts of oxygen needed for efficient nitrification. This is not surprising, as the system had limited time to develop; and may explain why NH_4-N production decreased over time in the planted tanks as reed growth accelerated, both through increased direct uptake of NH_4-N, and through a possible increase in nitrification near the developing root system not detected by the sampling regime. Sedimentation rates probably increased as the root mass developed, accounting for the decrease in the proportion of particulate N in the sewage. Increasing uptake by the developing biofilm may also be important, but was unquantifiable in this study.

It is possible that nitrification was occurring near to the roots, then rapidly denitrifying to the atmosphere, hence not being detected through the

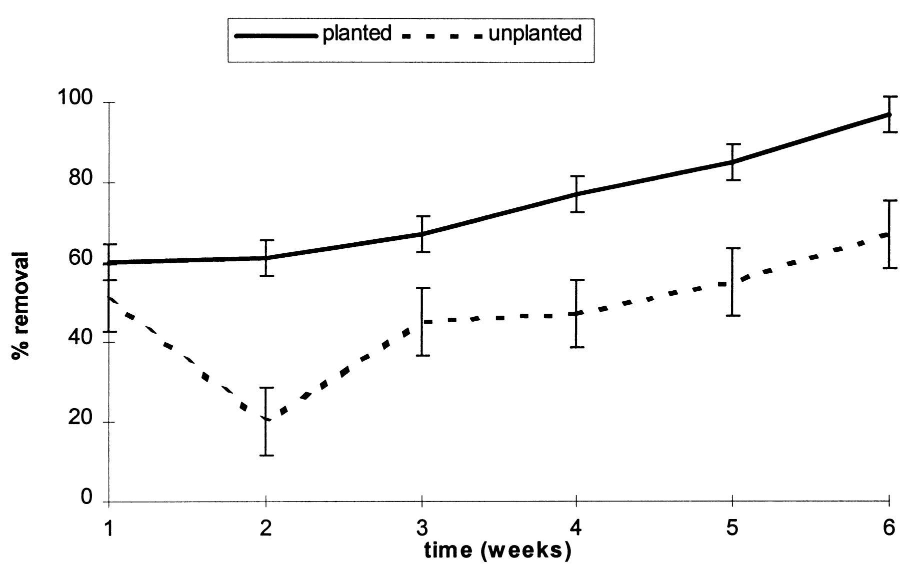

Fig. 1. Percentage removal of chemical oxygen demand over time. Error bars indicate standard deviation of three replicates.

sampling procedure. The pH, temperature and N:P ratio of the sewage were comparable to the optimal required by nitrifying bacteria. To investigate whether nitrification was occurring, samples would need to be taken from close to the plant roots and measured for dissolved oxygen and TON, or used to estimate numbers of nitrifying bacteria present.

Phosphorus removal

Whilst it is recognised that N removal is largely dependent on the developmental state of the system, and thus was not expected to be fully developed in this study, P removal by the substrate was expected to be more efficient. In fact the tanks removed phosphorus from the influent sewage very effectively.

The average TP found in the synthetic sewage solution over seven weeks was 7.49 mg l^{-1}. Phosphorus was consistently removed from the effluent by both treatments, with no significant differences between planted and unplanted tanks ($P = 0.11$) or in performance over time ($P = 0.08$), both removing 98% of influent P (Table 3).

Of the total phosphorus found in the sewage 93% was in the form of soluble reactive phosphorus (SRP) (6.96 mg l^{-1}). Both treatments on average removed 99% of the influent SRP. As with TP there were no significant differences found between or within treatments ($P > 0.05$).

Figure 3 compares the concentrations of TP and SRP (μg l^{-1}) found in the samples from planted and unplanted tanks over seven weeks of measurement. It can be seen that the amount of SRP approximately mirrors the pattern of TP for both treatments. On average, in planted tanks, SRP was 37% of TP, whilst for unplanted tanks SRP was 57% of TP. This difference may have been accounted for by uptake by the biota. Unplanted tanks gave more variable results, but neither treatment showed any definite trends over time.

General discussion

Although results obtained by different experimental methods and design should be compared only tentatively, the performance shown by both treatments at removing total nitrogen was low (between 40 and 60%) when compared with figures in the literature for gravel wetlands (between 47 and 80%; Vandevenne, 1995; Breen, 1990) and conventional treatment systems (between 8 and 98%; Horan, 1990; Kadlec and Knight, 1996). It is unlikely that the system in the experiment described here realised its full potential at removing nitrogen from the influent sewage due to the short duration of the study.

However, it was shown that maerl was effective at removing phosphorus from sewage, and indeed performance was better than that of many substrates described in the literature. Phosphorus removal, at 98%, exceeded the highest figures quoted for gravel bed wetlands (94% (Breen, 1990) and 95% (Rogers *et al.*, 1990), and were comparable with figures quoted for the most effective novel wetland substrates (for example 98% for LECA (Maehlum *et al.*, 1995) and 100% for red mud (Lopez *et al.*, 1998)). The maerl also performed well in excess of conventional waste water treatment systems such as trickling filters and activated sludge plants (25% and 90%, respectively (Horan, 1990)).

Maerl was successful as a P adsorbing substrate due to its high surface area, providing increased contact time with the effluent and many sites for

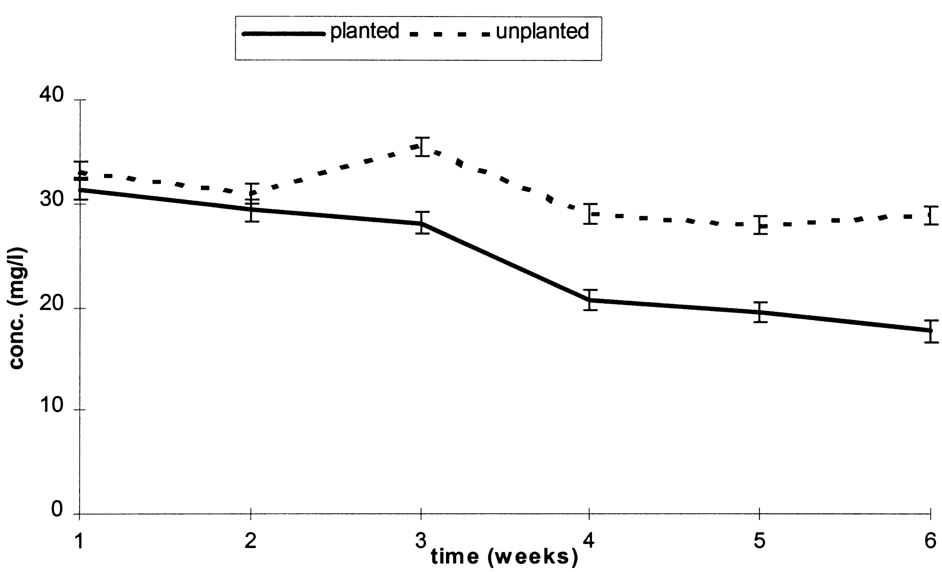

Fig. 2. Concentrations of NH_4-N in tanks (mg l^{-1}). Error bars indicate standard deviation of three replicates.

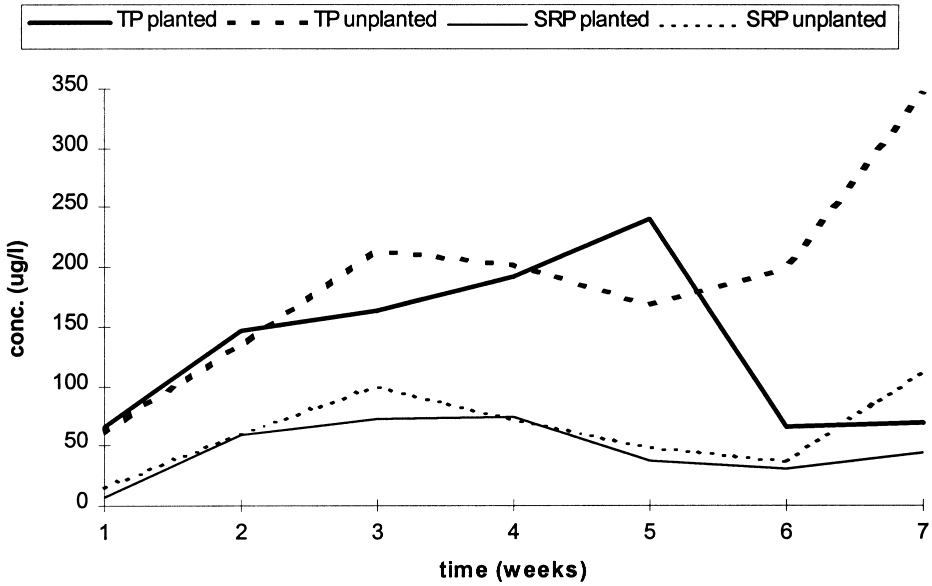

Fig. 3. TP and SRP concentrations ($\mu g\ l^{-1}$) in the tanks.

adsorption. The high calcium carbonate content enabled the chemical precipitation and subsequent sedimentation of phosphate. It is also believed that in a full-scale system, the trace elements present in the maerl could enhance plant and microbial growth, thus indirectly increasing uptake. However, as high temperatures increase the adsorption capacity of a substrate (Sakadevan and Bavor, 1998), and the temperature in the tanks was equivalent to a hot summer day, the efficiency of the substrate may actually have been artificially enhanced. The results of this study do indicate that maerl is worthy of further consideration as a constructed wetland substrate; especially where phosphorus removal is a priority. There are conservation issues associated with the extraction of maerl, and there is currently one license held in the UK for the extraction of 4000 T per year; believed to be a sustainable rate of harvest.

CONCLUSIONS

The main conclusions that can be drawn from this experiment are:

1. maerl is able to support the growth of wetland plants and the associated ecosystem, at least in the short term;
2. removal of P from effluent by maerl is highly effective as part of a wetland system. Nitrogen removal was low, with significant production of ammonium-N, but COD removal was high and improved over time. Although these results cannot be directly applied to the potential performance of maerl in a full-scale wetland, due to difficulties and problems peculiar to short-term indoor culture, this study gives an indication that the use of maerl should be the subject of further investigation in larger scale projects. Maerl could also be easily incorporated as a P filter in existing systems;
3. the equilibrium P-adsorption capacity of maerl is approximately 1200 mg kg^{-1}, giving the experimental tanks a lifetime of about 4 years at normal levels of P in sewage effluent;
4. the performance of maerl as a wetland substrate is comparable to figures quoted in the literature for other substrates and was higher in some instances.

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