# **The Role of Macrophytes in Nutrient Removal using Constructed Wetlands**

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## **1. Introduction**

#### **1.1 Overview**

This chapter reviews the role of aquatic plants (macrophytes) in the removal of nutrients from wastewater using constructed wetlands, with particular emphasis on surface-flow wetlands in tropical-subtropical climates. Nutrients (nitrogen and phosphorus) are potential contaminants in many wastewater effluent streams in urban, industrial and rural areas. This review focuses on the ecological requirements of macrophytes, the suitability of species for nutrient bioaccumulation and biomass production, and the overall performance and limitations of macrophytes in nutrient removal from the constructed wetlands.

 A recent review of the phytoremediation potential of wetland plants focussing on natural wetland ecosystems has been undertaken by Williams (2002) and is complementary to this review. Williams' review addresses the role of wetland plants in the phytoremediation of metals, volatile organic compounds, pesticides, herbicides, TNT (and other explosives) and hydrocarbons, but does not include nutrient removal.

 What are wetlands? Natural wetlands are areas that are permanently or periodically inundated or saturated by surface or groundwater and support the growth of aquatic vegetation (Mitsch and Gosselink 2000). Natural wetland types include saltwater wetlands (e.g. mangroves, salt marshes) and freshwater wetlands (e.g. sedgelands, reed beds, swamp forests and shallow lagoons). Wetlands are at the interface between terrestrial and aquatic environments and are strategically placed in the catchment, where they can intercept runoff water from uplands and floodwater from lowlands. Because of their strategic transitional location, floodplain wetlands are highly fertile areas. Globally, these natural wetlands have now disappeared due to cultivation for crops (Gopal 1999; Mitsch and Gosselink 2000).

 The use of specifically designed constructed wetlands for the treatment of wastewater (municipal, industrial, urban and agricultural) has been widely accepted over the past 20 years. However, the use of natural wetlands to assist in water purification has been in existence in many parts of the world for centuries. The functional processes were not understood until ecological research focused on the nutrient dynamics of wetland systems in the 1960s and 1970s. It is the interaction between abiotic and biotic components which are vital for water-quality improvement by either removing, recycling or storing contaminants (Reddy and D'Angelo 1997; Mitsch and Gosselink 2000; Wetzel 2001; Williams 2002). The plants and micro-organisms remove and recycle nutrients and metals either from the water or sediments. The sediments, biotic components and detritus (dead organic matter) are major storage components. For constructed wetlands to be effective in water pollution control, they must function as "pollutant" sinks for sediment, nutrients, metals, i.e. these pollutants must be removed from the wastewater and stored within the wetland either in the sediment or the plants.

#### **1.2 Wetland Processes to Improve Water Quality**

Various processes of wetlands are improving water quality are summarised in Table 1. Thus, the effectiveness of water-quality improvement is dependent upon an array of complex and interacting processes which can broadly be classified into three categories - physical, biological and chemical. Most processes are facilitated by the wetland vegetation.

<b>Potential Pollutant</b>	Role of the Wetland
Suspended solids including biodegradable particulates (BOD)	Sedimentation is facilitated by the vegetation. The vegetation reduces water velocity and turbulence causing settlement. Finer particles adhere to the biofilm surface of the vegetation. The root system binds and stabilises deposited particulates. The leaf litter and vegetation reduce resuspension.
Nutrients - nitrogen and phosphorus	Direct uptake by plants and micro-organisms. Inorganic nutrients converted to organic biomass. Microbial processes facilitate the removal and transformation of nutrients, especially nitrogen removal.
Metals	Direct uptake by plants and micro-organisms. Microbial bioremediation of metals. Metals immobilised by adsorption onto sediments or by precipitation.
Hydrocarbons	Microbial hydrocarbon degradation
Pathogens	Natural UV disinfection. Natural biocontrol by microbial predators in the wetland ecosystem. Adsorption to fine particles and sedimentation. Natural death and decay.

**Table 1.** Role of wetlands in improving water quality

## *1.2.1 Physical Processes*

Emergent macrophyte vegetation decreases water velocity, enabling the sedimentation of particles. Both submerged and emergent macrophytes are particularly effective in removing finely graded particles which will adhere directly onto the plant surface. The vegetation also distributes the flow and reduces turbulence, thereby allowing settlement of particles. The root system binds and stabilises deposited particles.

## *1.2.2 Biological Processes*

Plants and photosynthetic micro-organisms remove soluble inorganic nutrients (ammonium, nitrite, nitrate, phosphate) and heavy metals by direct uptake. Rooted macrophytes remove these nutrients from the sediment, whereas submerged and floating macrophytes and algae remove the nutrients directly from the water column. These inorganic nutrients are assimilated and converted into organic matter (biomass) and rendered relatively unavailable until death and decay.

 Macrophytes and photosynthetic micro-organisms also improve overall water quality by producing oxygen during photosynthesis which diffuses into the water column. Emergent macrophytes transport oxygen down their stems into the roots where it diffuses into the sediment to produce an aerobic microenvironment around the root zone (rhizosphere).

 The interaction between macrophytes and microbes is essential for nitrogen removal. Microbial processes of significance for the removal and transformation of nitrogen are ammonification, nitrification and denitrification. Ammonification is a decomposition process whereby dead organic matter (proteins) is converted to amino acids and then ammonia. Ammonification occurs under both aerobic and anaerobic conditions. Ammonium ions can either be assimilated by plants or nitrified under aerobic conditions by nitrifying bacteria to nitrites and nitrates. Sediments being waterlogged are often anaerobic, and therefore nitrification cannot proceed and ammonium ions dominate. However, in aerobic micro-environments around the rhizosphere of macrophytes, nitrification occurs. These nitrates can then be taken up directly by the roots. The dead organic matter of macrophytes provides a carbon source for the heterotrophic denitrifying bacteria.

## *1.2.3 Chemical Processes*

Chemical processes facilitate the adsorption and desorption of phosphorus onto and from sediment particles. Diffusion of oxygen from the roots of emergent macrophytes maintains an oxidised sediment surface layer and microenvironment around the root zone. This modifies the sediment redox conditions facilitating aerobic microbial processes including nitrification.

### **1.3 Applications of Constructed Wetlands**

Because of the ability of constructed wetlands to remove, recycle, transform and/or immobilise a wide range of potential contaminants, there are an ever expanding number of applications of constructed wetland technology. The most widespread use of constructed wetlands is in the treatment of domestic and municipal wastewater. Constructed wetlands can provide secondary treatment (after primary treatment of screening, sedimentation) and final polishing, i.e. advanced or tertiary treatment (after activated sludge process, trickling filters and/or oxidation ditches). In recent years, constructed wetlands to treat urban stormwater from housing estates and shopping centres have been incorporated into the urban landscape. Constructed wetlands are also being designed to hold and treat runoff from major roads and highways. Wetlands have also been constructed to intercept crop runoff in agricultural areas, particularly where there are sensitive downstream aquatic ecosystems. Dairy farms, cattle feed lots, piggeries and poultry farms generate concentrated animal waste which can be treated by constructed wetlands. Aquaculture farms are also pre-treating their water through wetlands prior to discharge into streams and rivers.

 Industrial applications are also increasing, and many of these have recently been reported in the literature (IWA 2000). They include:

- mining (acid coal mine drainage with high concentrations of dissolved iron, manganese, aluminium and sulphate; metal-mine drainage from lead, zinc, silver, copper, nickel and uranium mines)
- food processing wastes (peeling, pre-cooking and processing fruit and vegetables; sugar production; poultry and meat processing)
- x petrochemicals (polishing of secondarily treated refinery wastewater, treatment of washdown runoff)
- pulp and paper-mill wastewater
- treatment of landfill leachate and wastewater sludges

### **1.4 Wetland Plants**

Vegetation is the most conspicuous feature of wetlands. Wetland plants are morphologically and physiologically adapted to seasonal and/or permanent water inundation (Mitsch and Gosselink 2000). Aquatic plants are usually herbaceous and are referred to as hydrophytes. In constructed wetland technology, these aquatic plants are termed "macrophytes" (IWA 2000; US EPA 1988, 2000). Woody shrubs and trees also dominate many natural forested wetlands, e.g. mangrove swamps, *Cypress* swamps, *Melaleuca* swamps, riparian wetlands (Mitsch and Gosselink 2000).

 Macrophytes can be classified according to their morphological form or functional type (Fig. 1). There are two broad functional types: (i) rooted plants, and (ii) non-rooted plants. Rooted plants are anchored in the sediment and remove nutrients for the interstitial pore water. Non-rooted plants are not anchored, and either float on the surface or are suspended in the water column.

Rooted plants can be further classified as:

- Emergent macrophytes, i.e. roots/rhizomes in the sediment, and emergent stems and leaves which rise above the water (e.g. reeds, bulrush, sedges). Rooted emergent macrophytes are restricted to shallow water from a few centimetres to a maximum depth of 1 m.
- Floating-leaved macrophytes, i.e. roots/rhizomes in the sediment, stems submerged and leaves floating on the water surface (e.g. water lilies). Maximum depth 1.5 m.
- Submerged macrophytes, i.e. stems and leaves submerged (e.g. *Potamogeton*, *Triglochin*, *Vallisneria*). The depth distribution of submerged plants is restricted by light and oxygen availability.
- Creepers or vines, i.e. anchoring roots in shallow sediment, floating stems and leaves, with adventitious roots in the water (e.g. *Bacopa monniera*, *Ipomopa aquatica*, *Ludwigia peploides*, *Persicaria strigosum*).
- Trees and shrubs, i.e. woody plants that dominate seasonally inundated swamp forests, riparian zones and floodplains (e.g. *Melaleuca*, *Salix*, *Alnus*).



**Fig. 1.** Forms of aquatic plants found in constructed wetlands

Non-rooted plants can be further classified as:

- Floating plants, i.e. surface leaves (and stems) with roots which hang down into the water (e.g. water hyacinth (*Eichornia*), duckweed (*Lemna*), *Azolla*).
- Submerged plants, i.e. stems and leaves below the water surface; adventitious roots may be present but non-attached (*Ceratophyllum*, *Hydrilla*).
- Creepers or vines, i.e. floating stems and leaves with adventitious roots in the water (e.g. *Ipomea aquatica*, *Paspalum distichum*).

#### **1.5 Types of Constructed Wetland Systems**

There are basically two types of constructed wetlands - Free Water Surface Systems (FWS) and Sub-surface Flow Systems (SSF).

 Freewater surface flow (FWS) wetlands resemble natural wetlands in appearance and are composed of shallow (20-50 cm) vegetated channels or basins and deeper (50 cm - 2 m) open-water ponds. Vegetated shallow areas are often referred to as marshes. Marshes are typically dominated by emergent macrophytes, i.e. plants with roots in the sediment and emergent stems and leaves (reeds, sedges, rushes). However, floating-leaved attached macrophytes, i.e. plants with roots in the sediment and floating leaves (water lilies), submerged macrophytes (pond weeds) and floating macrophytes (e.g. duckweed) are also found in these shallow wetlands. Deeper ponds support floating macrophytes or submerged macrophytes if there is sufficient light for growth. FWS wetlands are typically used to provide tertiary wastewater treatment after conventional secondary treatment involving trickling filters or oxidation ponds, which remove most of the organic pollutants. They are most suitable for mild temperate or tropical/subtropical climates where freezing of the water does not occur in winter and continuous aquatic plant growth can occur.

 An FWS consists of channels or free-form shallow basins with a natural or constructed base of clay or impervious geotechnical material to prevent seepage, and a layer of suitable substrate to support rooted emergent macrophytes. Water depth can vary to suit the plant species used; lagoon configurations can also support floating aquatic plants. Substantial areas of land may be required to establish successful FWS.

 Sub-surface flow (SSF) wetlands, also known as vegetated submerged bed systems (US EPA 2000) in the USA, and reed-bed or root-zone wastewater treatment systems (IWA 2000) in Europe, are gravel and/or soil/sand-filled trenches, channels or basins with no standing water, and support emergent vegetation. They are typically used in Europe to provide secondary treatment after screening and primary settlement. Because of the potential for clogging of the media, they are mostly used for small communities or single households. They are suitable for cold climates as microbial processes can still occur in the root zone in winter. The absence of standing water, however, precludes the use of many aquatic plant species; only emergent species can survive in the waterlogged gravel or soil media. There is generally a higher treatment performance efficiency per unit area of land. Therefore, less land is required for the construction of SSF as compared to an FWS wetland. However, there is a higher capital cost associated with media supply and maintenance, if clogging occurs.

 An SSF consists of trenches with impermeable liners and a substrate of gravel and/or soil supporting emergent macrophytes. The systems can be designed to allow the wastewater to flow horizontally through the root zone, maximising filtration and sorption in the substrate, nutrient uptake by plants and micro-organisms and microbial degradation. Horizontal flow (HF) constructed wetlands are also termed reed bed treatment systems (RBTS) in Europe, because the reed *Phragmites australis* is commonly used. In North America, the term vegetated submerged bed (VSB) is used. Another type of SSF system is the vertical flow (VF) system. These layered gravel-sand reed beds are dosed intermittently with wastewater which is fed from the top, causing surface flooding. The wastewater drains vertically down, and the bed is then allowed to aerate before the next dosing.

 FWS systems are more suitable in subtropical/tropical conditions where year-round plant growth occurs. A wide range of plant species can be used (Greenway 2003). SSF systems are most prevalent in Europe and temperate regions of North America. Although the plants die back in winter, microbial activity continues. The range of suitable plants species for SSF systems is limited *Phragmites australis, Phalaris arundinacea, Glyceria maxima, Typha*  spp. and *Scripus* spp. have been used in Europe and North America (IWA 2000; Kvet et al*.* 1999). In sub-tropical Australia, *Baumea articulata, Carex fasicularis, Phylidrum languinosum* and *Schoenoplectus mucronata* are being trialled (Browning and Greenway 2003). Bolton used *Melaleuca* tree species (Greenway and Bolton 2002).

 The choice of plant species depends upon the physical structure of the constructed wetland which is governed primarily by the type of wetland system (FWS or SSF) and the pollutant characteristics, i.e. chemical composition of the wastewater effluent. The type of wetland system is determined by the extent of treatment required, i.e. secondary or tertiary wastewater treatment, the mass loading, climatic conditions, area of land available and cost (Greenway 2004; IWA 2000).

## **1.6 Ecological Requirements of Macrophytes in Treatment Constructed Wetland Systems**

Water, light, nutrients and oxygen are essential resources for the plant growth. Water and nutrients are the products associated with wastewater, in particular, sewage effluent, animal husbandry (piggeries, dairies), food processing, agricultural and urban stormwater runoff. A summary of the North American Treatment Database (IWA 2000) found average municipal wastewater effluent concentrations entering constructed wetlands were 3.8 mg/L PO<sub>4</sub>-P, 5.49 mg/L

 $NO<sub>x</sub>-N$ , and 4.97 mg/L NH<sub>4</sub>-N. Thus constructed wetlands are ideal candidates for promoting plant growth. However, at high concentrations, some nutrients, especially ammonium and phosphate, may become toxic to plant growth. For example, concentrations of  $NH_4-N$  in animal wastewater are extremely high, with average concentrations of 105 mg/L for dairy, 74 mg/L for poultry, and 366 mg/L for piggery (CH2M HILL and Payne Engineering 1997). Not only are such high concentrations of ammonium toxic to most macrophytes, but rapid oxygen depletion occurs due to nitrification.

 Water depth determines the different functional types of macrophytes found in constructed wetlands (Fig. 1). In natural wetlands, the distribution of the types and species of aquatic plants is usually governed by the water depth. Zonation is common with emergent, seasonally inundated species occurring at the landward interface, and submerged species or water lilies occurring in deeper permanent water. Free-floating species occur where there is open water, regardless of the water depth. Light is essential for photosynthesis and can limit the growth of submerged species where light penetration is reduced either through turbidity, shading, or very deep water. Oxygen is essential for aerobic respiration, and aquatic plants have morphological, anatomical and physiological adaptations for coping with the relatively low concentrations of dissolved oxygen in the water column and sediment. Emergent species can transport oxygen through special air spaces in their leaves and stems to the roots and rhizomes in the sediment. However, submerged species are unable to survive under anaerobic conditions. The distribution of species is also affected by substrate type and water quality (pH, salinity, toxic contaminants).

 In FWS treatment, wetlands water depth does not generally fluctuate, and is maintained between 20-50 cm depth. Water quality is often high in TSS, BOD and nutrients, and the sediment can become very anaerobic. Thus, the physicochemical conditions in treatment wetlands can be very different from natural wetlands. In treatment wetland systems, the plants need to be adapted to permanent waterlogging, and able to tolerate high nutrient concentrations in the water and sediment (Greenway 2003).

 The layout or configuration of wetland zones is important for treating all forms of wastewater (IWA 2000; Greenway 2004). Deep ponds or lagoons are appropriate as retention basins for stormwater wetlands or for treating wastewater effluent using floating plants, such as duckweed or water hyacinth. Large-scale treatment systems using floating plants require regular harvesting. Harvesting not only removes bioaccumulated nutrients (and metals), but also provides a potential resource as fodder for cattle or other livestock. Treatment lagoons can also function as aquaculture ponds for fish.

 Emergent macrophytes are an essential component of most constructed wetlands and play a major role in facilitating physical and biological processes in pollutant removal (Table 1). Emergent macrophytes, however, are restricted to shallower water, usually less than 50 cm deep, and not all species can tolerate permanent flooding.

 Surface Flow Wetland Systems for the treatment of steady-flow wastewater streams exhibit a little variation in water levels and are usually designed for a depth of 30-50 cm of water. Thus, sedges and reeds tolerant of permanent inundation need to be planted. By contrast, huge fluctuations in water levels occur in stormwater wetlands, necessitating a range of shallow (< 10 cm) and deeper (50 cm) macrophyte zones. The shallower zones will completely dry out during low rainfall periods. Therefore, plant species, that can tolerate a wetting and drying cycle, should be selected for these areas. A diversity of vegetation zones can also enhance the overall wildlife value of the wetland as well as the landscape amenity.

## **2. Role of Macrophytes in Nutrient Removal**

## **2.1 Overview**

One of the primary factors that has attributed to the use of constructed wetland systems for municipal wastewater treatment is "recognition of the natural treatment functions of aquatic plant systems and wetlands, particularly as nutrient sinks and buffering zones" (US EPA Design Manual 1988). As outlined in Section "Wetland processes to improve water quality", nutrient removal, transformation, recycling and retention are largely biologically mediated. The macrophytes either directly or indirectly play an important role in nutrient removal and storage. The removal of soluble inorganic nitrogen and phosphorus via absorption from either the water column or the sediment, assimilation and storage in plant tissue, is a direct mechanism of nutrient sequestration. The provision of plant surfaces (leaves, stems and roots) for attached microbiota, epiphytic microflora and associated biofilm communities enables microbial assimilation, transformation and storage of nutrients. Although there is still debate about the relative importance of macrophytes versus microbes in nutrient removal (Brix 1997; IWA 2000; Tanner 2001; Wetzel 2001), plant biomass still accounts for substantial removal and storage of N and P (Rejmankova et al. 1990; IWA 2000; Greenway and Woolley 2001).

 Since inorganic nitrogen and phosphorus are essential for the plant growth, it is possible to maximise the amount of nutrients removed from wastewater effluent by selecting macrophytes with a high capacity for inorganic nutrient absorption and conversion to organic plant biomass. They should have a long or continuous growing season and be highly productive and capable of accumulating large amounts of nutrients in plant biomass. Rooted plants remove nutrients directly from the sediments, whereas floating plants remove nutrients from the water column. Some emergent species, such as *Phragmites,* have adventitious "water roots", and the water snow flake *Nymphoides* produces roots from the floating leaf base, thereby enabling these species to remove nutrients from both sources. Many submerged species obtain nutrients directly from the water column via leaf absorption, particularly in species with poorly developed root systems, such as *Ceratophyllum*. Once nutrients have been absorbed, they can be translocated to other parts of the plant. Below-ground storage in rhizomes is common in emergent macrophytes.

 Plant uptake is an important nutrient removal mechanism in wastewater treatment systems. Contact between the active zones of nutrient absorption and the wastewater or sediment must be maximised to optimise nutrient removal and incorporation into plant biomass.

#### **2.2 Suitability of Macrophyte Species**

In North America, 851 wetland plant species have been identified (Knight et al. 2001), of which 593 species have been recorded in constructed treatment wetlands. In subtropical-tropical Queensland, Australia, 150 wetland plant species have been identified as "potential aquatic plants for use in freewater surface flow constructed wetlands" (QDNR 2000), of which 72 have been found growing (planted or self-colonised) in treatment wetlands (Table 2) (Greenway 2003).

 These two examples demonstrated the huge potential of using aquatic plant species which occur naturally in wetlands or waterways. However, as discussed earlier, for successful growth, the species selected must be able to tolerate permanent waterlogging, higher nutrient concentrations, lower dissolved oxygen due to high BOD (and COD) loads, higher turbidity due to high TSS loads, and potentially toxic contaminants depending on the source of the wastewater.

 Greenway (2003) found that all species listed in Table 2 were able to grow successfully in secondary-treated sewage effluent with  $PQ_4$ -P concentrations 2.5-8.7 mg/L,  $NO_3$ -N concentrations 9.7-15.8 mg/L, and  $NH_4$ -N concentrations 7.7-18.6 mg/L. The lowest species richness, however, occurred in a wetland receiving effluent with 22-30 mg/L NH4-N. *Typha domingensis*, *Ludwigia peruviana*, the aquatic creepers *Ludwigia peploides*, *Paspalum distichum* and *Persicaria orientalis*, and duckweed were the only species to spread successfully. *Typha* was planted, but the other species were natural invaders and colonisers.

 While Table 2 provides a list of species suitable for surface-flow wetlands in tropical-subtropical Australia, most of these genera and several species are cosmopolitan in distribution.

 Many macrophyte species have been trialled successfully for use in SSF CW around the world (Tables 3 and 4). The most widespread and commonly used emergent species is the reed *Phragmites australis*. Species used in Europe include *Phalaris arundinaceae* (reed canary grass), *Glyceria maxima* (sweet manna grass) and *Typha* spp., and in the USA *Scirpus* spp. (IWA 2000). Commonly used species in Australia and New Zealand include *Phragmites*, *Schoenoplectus* and *Juncus* spp. (Browning and Greenway 2003).

Family	Species and Genus
Alismataceae	*Sagittaria graminea (E)
Apiaceae	Hydrocotyle bonariensis (FF)
Amaranthaceae	**Alternanthera philoxeroides (E or FF)
Araceae	Pistia stratiotes (FF), *Colocasia esculenta (E)
Asteraceae	Eclipta prostrata (E or $FF$ )
Azollaceae (fern)	Azolla sp (FF)
Cannaceae	*Canna sp. $(E)$
Ceratophyllaceae	Ceratophyllum demersum (S)
Convolvulaceae	Ipomoea aquatica, Ipomoea diamantinensis (FF)
Cyperaceae	Baumea articulata, B. rubiginosa, Bolboschoenus fluviatilis,
(all emergents)	B. caldwelli, Cyperus alopercuroides, C. eragrostis,
	C. exaltatus, *C. papyrus, *Cyperus involucratus, Eleocharis
	acuta, E. dulcis, E. phillippinensis, E. sphacelata,
	Rhynchospora corymbosa, Scirpus sp., Scheonoplectus
	mucronatus, S. validus, Scleria poiformis, Schoenus apogon
Gramineae	*Brachiara mutica, *Echinochloa crus-galli, *E. colona,
(all emergents)	E. polystachya, Hymenachne acutigluma, *H. amplexicaulis,
	Leersia hexandra, *Pennisetum alopercuroides, Phragmites
	australis, Paspalum distichum (FF)
Hydrocharitaceae	Vallisneria gigantea (S)
Juncaginaceae	Triglochin procera (S)
Juncaceae	Juncus planifolius, J. polyanthemus, J. prismatocarpus,
	J. kraussii, J. usitatus (all E)
Lemnaceae	Lemna spp., Spirodela spp., Wolffia spp. (all FF)
Limnocharitaceae	*Hydrocleys nymphoides (FL)
Marantaceae	*Thalia dealbata (E)
Marsileaceae (fern)	Marsilea mutica (FL), Marsilea drummondii (FL)
Menyanthaceae	Nymphoides indica ((FL)
Myrtaceae	Melaleuca quinquenervia $(T)$
Nymphaeaceae	Nymphaea capensis (FL), Nymphea gigantea (FL)
Onagraceae	Ludwigia peploides (FL or FF), *Ludwigia peruviana (E),
	L. octovalvis $(E)$
Parkeriaceae (fern)	Ceratopteris thalictroides (FF or $E$ )
Philydraceae	Philydrum lanuginosum (E)
Polygonaceae	Persicaria attenuata, *P. orientale, P. strigosum (E or FF)
Pontederiaceae	Monochoria cyanea (FF)
Potamogetonaceae	Potamogeton crispus, P. pectinalis $(S)$
Salvinaceae (fern)	**Salvinia molesta (FF)
Scrophulariaceae	Bacopa monnieri (FF)
Typhaceae	Typha domingensis $(E)$ , Typha orientalis $(E)$
	$E =$ emergent, $S =$ submerged. FL = floating leaved attached. FF = free-floating or

**Table 2.** Macrophyte species occurring in constructed surface-flow tertiary-treatment wetlands in Queensland, Australia

 $E =$  emergent,  $S =$  submerged,  $FL =$  floating leaved attached,  $FF =$  free-floating or aquatic creepers,  $T =$  tree. \*Exotic (including introduced naturalised species), \*\*Noxious weeds in Australia

Family	Species and Genus
Alismataceae	Sagittaria, Sagittifolia
Cyperaceae	Carex spp., Rhynchospora spp., Cyperus spp., Scirpus spp.,
	S. acutus, S californicus, S. lacustris, Schoenoplectus validus,
	S. tabernaemontoni
Gramineae	Glyceria maxima, Panicum spp., Phalaris arundinacea,
	Phragmites australis, P. communis
Iridaceae	Acornus calamus, Iris pseudacorus, I. versicolor
Juncaceae	Juncus spp., J. effusus
Lythraceae	Lythrum spp., L. salicaria
Onagraceae	Epilobium spp.
Polygonaceae	Polygonum amphibium, Rumex spp., R. crispus
Sparganiaceae	Sparganium erectum, S. americanum
Typhaceae	Typha spp., T. latifolia, T. angustifolia, T. domingensis
Umbelliferae	Oenanthe spp.

**Table 3.** Macrophyte species used in subsurface-flow wetlands in temperate climates

**Table 4.** Macrophyte species used in subsurface-flow (SSF) wetlands in subtropical/tropical climates (Americas, Southern Asia, Australasia, Africa)

Family	Species and Genus
Cyperaceae	Carex fasicularis, Cyperus articulatus, C. flabelliformis, C. immensus, C. papyrus, Schoenoplectus validus, Scirpus californicus, S. lacustris
Graminaceae	Miscanthidium violaceum, Paspalum penisetum, Phragmites spp., P. australis, P. karka, P. mauritianus, Vetiveria zizanioides, Zizaniopsis bonariensis
Typhaceae	Typha spp., T. dominguensis, T. latifolia, T. subulata

#### **2.3 Nutrient Bioaccumulation**

One of the many roles macrophytes play in CW for wastewater treatment includes their capacity for nutrient bioaccumulation, i.e. the direct uptake and storage of nutrients (Brix 1997). Desirable plant characteristics for macrophyte species to maximise nutrient uptake in a constructed wetland treating secondary effluent, include rapid growth, high plant-tissue content, and the ability to attain a high standing crop (Reddy and De Busk 1987; Greenway 2003). Additional to these characteristics is the ability to recover following cropping, and an attractive species is also desirable to increase wetland aesthetics.

 The rate of nutrient uptake by macrophytes is limited by its growth rate and the concentration of nutrient within the plant tissues, with nutrient storage dependent on plant-tissue nutrient concentrations and potential for biomass accumulation (maximum standing crop) (Reddy and De Busk 1987). At low to medium nutrient concentrations, plant growth (biomass) is proportional to

nutrient supply (Fig. 2). Increases in nutrients above this may result in the luxury uptake of nutrients by plants, but does not increase plant growth. Nutrient accumulation will eventually plateau. Beyond this point, increases in nutrient supply may cause nutrient toxicity.

 A few plant species have been only grown successfully in constructed wetlands receiving high ammonium concentrations. Hunt and Poach (2001) stated that *Scirpus*, *Typha* and *Juncus* were the most commonly used plant genera in animal wastewater treatment. However, Kantawanichkul et al*.* (2003) reported retarded growth of *Scirpus grossus* in experimental tanks after 120 days.



**Fig. 2.** Relationship between nutrient supply and nutrient accumulation  $(- - )$  and biomass yield ( $\qquad$ ) (after Reddy and DeBusk 1987)

 Nutrient content also changes with the plant and leaf age. Young plants and leaves often have the highest nutrient content, especially nitrogen. As the plant (or the leaf) reaches maturity, its nutrient content decreases. However, since plant biomass increases with maturity, total nutrient storage (bioaccumulation) also increases. Upon senescence, nutrients from the mature leaves are translocated to the growing shoots or storage organs. Thus, dead shoots have less nitrogen and phosphorus levels. Harvesting dead shoots will, therefore, not optimise nutrient removal.

 A study by Greenway and Woolley (1999) of seven municipal wastewater treatment wetlands found no significant difference in the nutrient content of plants sampled from the inlet and outlet zones, suggesting nutrient sufficiency even in the final effluent.

 In a comparative study of the nutrient content of plants of the same species collected from constructed wetlands and natural waterways, Greenway (1997) found that the plants growing in constructed treatment wetlands had a higher N and P tissue content (Table 5). For most pairs of species, the statistical difference was not significant for nitrogen content, but phosphorus content in the treatment plants was almost double for many species. The variability among control plants may have been due to higher nutrient concentrations even in the natural waterways, particularly nitrogen. These results, however, demonstrate the capacity for increased nutrient accumulation with increased nutrient supply (Fig. 2).

**Table 5.** Comparison of P and N content (mg/g dry wt) in leaf/stem tissue in selected macrophyte species in constructed (treatment) and natural (control) wetlands. (mean  $\pm$ SD)

Species	Type	Treatment		Control	
		P	N	P	N
Phragmites australis	E	$2.0 \pm 0.6$	$20.4 \pm 8.0$	$1.4 \pm 0.6$	$12.0 \pm 7.3$
Typha domingensis	E	$2.3 \pm 1.0$	$15.8 \pm 6.0$	$1.5 \pm 0.9$	$9.2 \pm 6.2$
Baumea articulata	E	$3.7 \pm 1.9$	$13.1 \pm 4.2$	$2.4 \pm 0.7$	$12.6 \pm 2.4$
Schoenoplectus validus	E	$2.6 \pm 1.2$	$14.6 \pm 5.0$	$0.8 \pm 0.5$	$10.5 \pm 5.0$
Eleocharis sphacelata	E	$2.7 \pm 1.1$	$15.8 \pm 4.0$	$1.3 \pm 0.6$	$11.3 \pm 5.6$
Ludwigia peploides	FF	$5.4 \pm 1.8$	$36.9 \pm 10.2$	$3.1 \pm 1.0$	$27.0 \pm 10.2$
Persicaria orientalis	E	$4.4 \pm 1.3$	$33.2 \pm 10.1$	$1.4 \pm 0.6$	$13.5 \pm 3.3$
Nymphoides indica	FL.	$6.6 \pm 2.0$	$25.8 \pm 11$	$2.5 \pm 0.6$	$22.1 \pm 3.6$
Nymphaea gigantea	FL	$4.2 \pm 2.0$	$28 \pm 10$	$2.2 \pm 0.5$	$22 \pm 8$
Paspalum distichum	FF	$2.8 \pm 1.2$	$12.5 \pm 3$	$0.9 \pm 0.4$	$11 \pm 2.6$
Marsilea mutica	FL.	$8.0 \pm 1.6$	$28.8 \pm 3$	$2.2 \pm 0.5$	$14.7 \pm 3.4$

After: Greenway 1997

 Greenway and Bolton (2002) and Bolton and Greenway (1997, 1999) also found that the leaves of *Melaleuca* trees growing in sewage effluent had a significantly higher P content than the leaves and trees growing in a natural *Melaleuca* wetland. However, in P-enriched effluent  $(12 \text{ mg } P L^{-1})$ , decreased growth rates indicated P toxicity, with senescent leaves having the highest P content.

#### *2.3.1 Nutrient Content of Plant Components*

A comparison of phosphorus and nitrogen in both root/rhizomes and leaf/stem tissue for a varie2ty of macrophytes has been shown in Table 6. Nitrogen content is highest in the leaves, and phosphorus in the root/rhizomes. From Tables 5 and 6, it can be seen that the highest nutrient content occurs in duckweed, followed by *Ludwigia peploides, Ceratopteris, Monocharia, Bacopa, Ipomoea, Ceratophyllum, Nymphaea* and *Nymphoides* - all these species remove nutrients from the water column. There was not a large variation in the mean values of the emergent macrophytes.

Species	Root/Rhizome		Leaf/Stem			
	P N		P	N		
<b>Emergent macrophytes</b>						
Baumea articulata	$3.5 \pm 1.0$	$20.0 \pm 4.2$	$1.9 \pm 0.5$	$16.3 \pm 3.8$		
Bolboschoenus caldwellii	$4.3 \pm 1.5$	$13.5 \pm 5.0$	$3.0 \pm 1.4$	$14.3 \pm 5.4$		
Cyperus eragrostis	$3.7 \pm 1.8$	$20.0 \pm 9.6$	$3.7 \pm 1.7$	$16.5 \pm 4.2$		
Cyperus exaltatus	$5.0 \pm 4.0$	$15.0 \pm 7.0$	$3.8 \pm 1.8$	$16.7 \pm 6.8$		
Eleocharis acuta	$4.0 \pm 2.7$	$14.0 \pm 5.0$	$3.4 \pm 1.5$	$18.8 \pm 5.4$		
Eleocharis phillipensis	$4.4 \pm 2.0$	$14.0 \pm 4.8$	$3.5 \pm 1.2$	$17.2 \pm 4.9$		
Eleocharis sphacelata	$4.3 \pm 2.5$	$13.5 \pm 5.7$	$2.7 \pm 1.0$	$15.8 \pm 4.0$		
Rhynochosporus corymbosa	$2.5 \pm 1.9$	$13.9 \pm 0.6$	$2.5 \pm 0.6$	$16.9 \pm 2.4$		
Scheonoplectus validus	$4.0 \pm 1.9$	$14.5 \pm 7.0$	$3.9 \pm 1.3$	$18.2 \pm 4.1$		
Scleria poiformis	$2.8 \pm 0.8$	$12.3 \pm 2.5$	$2.6 \pm 1.0$	$16.0 \pm 4.0$		
Phragmites australis	$3.2 \pm 1.4$	$17.3 \pm 7.0$	$2.0 \pm 0.6$	$20.4 \pm 8.0$		
Typha sp.	$4.0 \pm 1.7$	$16.8 \pm 10$	$2.3 \pm 0.8$	$15.8 \pm 6.0$		
<b>Floating-leaved attached</b>						
Nymphaea sp.	$7.1 \pm 0.7$	$30.3 \pm 1.7$	$4.0 \pm 1.0$	$30 \pm 8.6$		
Nymphoides indica	$12.1 \pm 3.7$	$19.8 \pm 6.3$	$6.6 \pm 2.0$	$25.8 \pm 11$		
<b>Free-floating macrophytes</b>						
Azolla sp.	Whole plant		$7.4 \pm 1.0$	$40.0 \pm 4.0$		
Duckweed (Spirodela sp.)	Whole plant		$12.4 \pm 4.1$	$39.6 \pm 10.3$		
Ceratopteris thalicoides	Whole plant		$8.3 \pm 1.0$	$30.0 \pm 9.0$		
Monocharia cyanea	Whole plant		$7.7 \pm 3.1$	$20.9 \pm 9.0$		
<b>Submerged macrophytes</b>						
Ceratophyllum demersum	Whole plant		$15.4 \pm 4.4$	$27.0 \pm 6.3$		
<b>Vines/creepers</b>						
Bacopa monnieri	Whole plant		$4.8 \pm 1.1$	$30.0 \pm 11.2$		
Ipomoea aquatica	Whole plant		$6.4 \pm 1.2$	$30.7 \pm 12.4$		
Ipomoea diamentinenis	Whole plant		$7.5 \pm 2.1$	$37.3 \pm 11.9$		
Ludwigia peploides	Whole plant		$5.3 \pm 2.3$	$32.7 \pm 12.3$		
Persicaria attenuatum	Whole plant		$4.2 \pm 1.7$	$24.9 \pm 11.4$		
Paspalum distichum	Whole plant		$4.1 \pm 2.0$	$18.6 \pm 11.0$		

**Table 6.** Phosphorus and nitrogen contents (mg/g dry wt) (mean  $\pm$  SD) in plant parts of native macrophytes in FWS wetlands in Queensland, Australia.

#### **2.4 Biomass Production**

#### *2.4.1 Plant Biomass*

The rate of removal of nitrogen and phosphorus by plants and the incorporation of these nutrients into plant biomass are important for accessing the suitability of macrophyte species for phytoremediation. The turnover rates for plant biomass, individual plants, leaves/stems, and the nutrient storage capacity also need to be considered. The harvesting potential of plant biomass and the rate of regrowth following cropping are important for permanent removal of nutrients from the system.

 Emergent macrophytes, in particular *Phragmites* and *Typha*, have very high plant biomass (IWA 2000). Vymazal et al*.* (1999) reviewed the literature on *Phragmites australis* and found that natural stands of *Phragmites*, growing in eutrophic waters, can achieve a total biomass of  $12,700$  g/m<sup>2</sup>. They suggested that in constructed wetlands, high organic loads might stress the plants. However, the nutrient contents of plants from both natural stands and constructed wetlands were comparable.

 Tables 7 and 8 provide information on whole plant biomass (shoots, rhizomes/roots) of selected macrophytes growing in a surface-flow constructed wetland in Cairns, tropical Australia (Greenway and Woolley 1999 2001). Biomass for floating macrophytes - duckweed (*Spirodela* and *Wolffia*) and *Azolla* was determined in sections of open water and where the plants grew among emergents. Biomass standing stock of duckweed in open water was 100% higher.

 Biomass production values can be used as an indicator to estimate the nutrient uptake capacity of the plants. The uptake capacity of emergent macrophytes is in the range of 30 to 150 kg P/ha/y and 200 to 2500 kg N/ha/y (Brix 1997). Turnover rates for floating macrophytes, however, can be in the order of days or weeks. Under optimal conditions, some species of duckweed can double their biomass in 24 hours (Landolt 1996). Rejmankova et al*.* (1990) predicted optimum growth of 5.9  $g/m^2/d$  by removing 25% of duckweed cover every four days and 2.1  $g/m^2$  by removing 75% of cover. Doubling times of  $7-12$  days have also been reported in temperate  $(10-15^{\circ}C)$ wastewater ponds (Ozimek 1996). Without any harvesting, however, optimal growth is unlikely.

 By comparing the mass removal of N and P from the effluent with the N and P content in plant biomass, it is possible to get some indication of how much N and P may have been removed directly by the plants themselves. In a three-year study, Greenway and Woolley (2001) found that the uptake capacity ranged from 134 to 162 kg P/ha/y and 380 to 474 kg N/ha/y. This represented between 67 and 80% of  $PO_4$  removal, and 65-80% of soluble inorganic N removal which had been incorporated into plant biomass.

Species	<b>Biomass</b>	P Content	P Storage	N Content	N Storage
	$g/m^2$	mg P/g	g P/m <sup>2</sup>	mg N/g	g N/m <sup>2</sup>
	$mean \pm SD$				
Typha orientalis					
1 shoot	$125 \pm 75$	3.85	$0.48$ /shoot	13.5	$1.69$ /shoot
14 shoots/ $m^2$	$1750 \pm 750$		6.74		23.63
Eleocharis					
Dense	$1000 \pm 250$	4.2	4.20	15	15.0
mid dense	$500 \pm 300$	4.2	2.10	15	7.5
sparse	$300 \pm 140$	4.2	1.26	15	4.5
Schoenoplectus	$800 \pm 500$	3.5	2.80	14.5	11.6
validus					
Schoenoplectus	$360 \pm 390$	3.5	1.01	14.5	5.22
validus					
(among Typha)					
Marsilea spp	270 young	9.5	2.57	27	7.29
	370 mature	9.5	3.52	27	9.99
	470 old	7.3	3.42	19	9.04
Nymphoides indica	$83 \pm 20$	8.2	0.68	22	1.83
Paspalum	$860 \pm 110$	2.8	2.39	16	13.30
distichum					
Alternathera	$780 \pm 170$	3.2	2.50	16	12.48
philoxeroides					
Duckweed (open	$40 \pm 10$	14.4	0.58	43	1.72
water)					
Azolla and	$33 \pm 7$	8.0	0.26	41	1.36
Duckweed					
(open water)					
Duckweed (among	$20 \pm 4$	14.4	0.29	43	0.86
emergents)					
Azolla and	16	8	0.13	41	0.66
Duckweed					
(among emergents)			1.71	31	2.74
Ceratophyllum	$90 \pm 30$	18.95			
demersum					

**Table 7.** Plant biomass (g dry  $wt/m^2$ ), nutrient content of plant tissue (mg/g) and nutrient storage  $(g/m^2)$  in selected macrophytes (source: Greenway and Woolley 2001)

 Plant biomass and annual production rates are high in the tropical/subtropical regions (Greenway and Woolley 2001; Browning and Greenway 2003). In an experimental mesocosm band planted with *Phragmites*, *Schoenoplectus* and *Eleocharis*, mean shoot biomass after 12 months growth was 2200 g DW/m<sup>2</sup> (7 g P; 38 g N) (QDNR 2000). Regrowth from cropping yielded an annual production rate of 3500  $g/m^2$  and 10 g P; 58 g N. These growth rates are comparable to the Cairns study (Greenway and Woolley 2001), and further indicate the potential of these macrophyte species for nutrient



Species	<b>Whole Plant Biomass</b> $(g/m^2/y)$			Harvestable Shoot <b>Biomass</b> $(g/m^2/y)$		
	g dry wt	g N	g P	g dry wt	g N	g P
<b>Surface flow</b>						
Typha orientalis	4000	53.7	15.3	2264	30.6	8.7
Eleocharis sphacelata	3210	46.8	13.1	918	13.8	3.9
Schoenoplectus validus	1000	14.5	3.5	581	8.4	2.0
<b>Subsurface flow</b>						
<i>Phragmites australis</i>	6700	127.0	16.1	3564	71.0	6.7
Baumea articulata	3470	55.5	9.0	2512	41.0	4.8
Carex fasicularis	3920	60.0	9.1	2424	41.0	6.5
Schoenoplectus mucronatus	890	15.4	3.3	842	15.3	3.3
Philydrum lanuginosum	1000	17.1	2.5	947	16.9	2.2

**Table 8.** Biomass production (growth rate  $g/m^2/y$ ) after harvesting shoots in selected macrophytes from a tropical surface-flow wetland and a subtropical subsurface-flow wetland

(After: Greenway and Woolley 2001; Greenway 2002)

removal. In New Zealand, Tanner (2001) reported maximum nutrient accumulations of 8.8-13.4 g  $P/m^2$  and 48-69 g N/m<sup>2</sup> in total biomass (shoots and below ground) in *Schoenoplectus*, but due to the higher nutrient loading rates compared to the Cairns studies, this only accounted for 2-8% total N removal from the system. Tanner concluded that "uptake and storage of N and P in live plant biomass can usually only account for a fraction of the improved performance of the planted systems". However, in assessing the importance of macrophytes in nutrient removal, it should be recognised that plants have a maximum removal and storage capacity (Fig. 2). Thus, if effluent loading rates are low, then the relative removal efficiency by macrophytes will be higher than constructed wetlands receiving high nutrient loading rates.

# **3. Conclusion**

Vegetation is the dominant feature of constructed wetlands; the aquatic macrophytes and their associated microbial biofilms play several vital roles in removing, transforming and storing nutrients. The stems and leaves reduce water velocity and turbulence, causing filtration and settlement of particles (sediment, organic particulates), and provide an increased surface area for the attachment of epiphytic algae and micro-organisms. Oxygen produced in photosynthesis aerates the water. Inorganic bioavailable nutrients for plant and algal growth are removed either from the water column or the sediments. Oxygen transfer from aerial stems to the roots is released into the rhizosphere, facilitating the nitrification/denitrification process. Thus, macrophytes play a major role either directly or indirectly in the removal of nutrients from wastewater.

 The performance efficiency of constructed wetlands depends on several variables - these include the quality and quantity of effluent to be treated; the extent of physical, biological and chemical processes functioning within the wetland; the contact time of wastewater with sites of biological and physical activity. Reactive biological surfaces include the plants and associated biofilms, the litter layer and/or sediment and associated microbial communities. The flows and storage volume determine the detention time (hydraulic retention time - HRT) and thus the opportunity for interactions between wastewater contaminants and the wetland ecosystem. In temperate climates, water temperature can influence biological processes. Nutrient removal can be optimised by using macrophytes with a high capacity for inorganic nitrogen and phosphorus absorption and conversion into plant biomass.

 However, it should be recognised that all plant species have a maximum nutrient uptake and storage capacity in plant biomass. Species should, therefore, be selected with high nutrient removal capabilities, which means a mixture of macrophyte types (submerged, floating and emergent species) should be used. In tropical/subtropical climates with continuous growing seasons, a mixture of emergent macrophytes, submerged and floating species (duckweed) in surfaceflow systems can contribute significantly to the removal of nitrogen and phosphorus from wastewater. However, to maximise removal efficiencies, effluent loading rates should not be too high. Harvesting of plant biomass may be suitable for floating species, such as duckweed, or water hyacinth, which can offer a resource benefit.

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