



Site test of phytoremediation of an open pond contaminated with domestic sewage using water hyacinth and water lettuce



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ABSTRACT

This study was undertaken *in situ* to explore the potential of the alien plants water hyacinth (*Eichhornia crassipes*) and water lettuce (*Pistia stratiotes* L.) as phytoremediation aquatic macrophytes for nutrients (nitrogen and phosphorus) removal and algal interception from domestic sewage contaminated pond (approximately 10500 m² in area, average 2.5 m in depth) by using self-designed experimental devices from July 7 to August 8 in 2015. The physicochemical properties of water and plant samples as well as N and P mass balance in the phytoremediation system were investigated. The range of physicochemical parameters of influent were shown as follows: water temperature (WT: 24.5 °C–31.0 °C), pH (6.94–8.25), DO (4.58 mg L⁻¹–15.73 mg L⁻¹), COD_{Mn} (5.00 mg L⁻¹–13.15 mg L⁻¹), TN (1.60 mg L⁻¹–5.60 mg L⁻¹) and TP (0.16 mg L⁻¹–0.73 mg L⁻¹). Water hyacinth, which exhibited hyperactive accumulating capacity for nitrogen (58.64% of total reductions), was more suitable than water lettuce for the intensive purification of domestic sewage with high nitrogen concentrations. This result may be attributed to the larger total root surface area (0.97 m² g⁻¹–1.10 m² g⁻¹ fresh weight), active absorption area (0.31 m² g⁻¹–0.36 m² g⁻¹ fresh weight), and leaf area and higher root activity (71.79 μg g⁻¹ h⁻¹–98.34 μg g⁻¹ h⁻¹), root biomass (kg m⁻²), and net photosynthetic rate (20.28 μmol CO₂ m⁻² S⁻¹) of water hyacinth than those of water lettuce regardless of cultivation in oligotrophic water with total nitrogen contents lower than 1.0 mg L⁻¹. Water lettuce exhibited a higher total phosphorus removal efficiency, which benefitted higher P accumulation, adsorption, and precipitation because of its longer roots (approximately 49.0 cm) with higher rhizofiltration capacity. As such, water lettuce achieved higher algal (96.36%) and chlorophyll *a* (96.65%) removal efficiencies. A combined pattern using both macrophytes was recommended for the phytoremediation of most domestic sewages containing dual contaminants (N and P) in the future.

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1. Introduction

In recent years, human-related eutrophication has been considered a common environmental issue and has received worldwide attention (Conley et al., 2009), especially for many developing countries. Among the various measures for eutrophication control, phytoremediation, a solar-driven biological method performed directly *in situ* (Salt et al., 1998), has received considerable attention because of its low cost and environmental friendliness (Batty and Dolan, 2013). Phytoremediation, in which plants are used to remediate a contaminated medium, is a well-established environmental protection technique that has received increasing attention since the term has been coined two decades ago (Vamerli et al., 2010;

Priya and Selvan, 2014). Plants used for phytoremediation must meet requirements, such as high biomass, rapid growth, and high nutrient accumulation; in addition, the application of this method is limited by its clean-up depth (purification depth from water surface), which is strictly determined by plant root length (Putra et al., 2015).

Water hyacinth (*Eichhornia crassipes* Mart. Solms.), a member of the monocotyledonous family Pontederiaceae (Patel, 2012), is a free-floating perennial aquatic macrophyte native to tropical South America (Tipping et al., 2011). The plant is notorious as a highly noxious alien weed because of its tremendously vigorous growth rate (Mishra and Tripathi, 2009). However, given its dense hairy root system, peculiar physiological characteristics and nutrient absorption efficiency (Kim and Kim, 2000; Paganetto et al., 2001), and wide tolerance to environmental conditions (Rommens et al., 2003), water hyacinth has been widely utilized and has gained increasing

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attention in recent years for the phytoremediation of many types of wastewater (Chavan and Dhulap, 2012).

The monocotyledonous freshwater floating macrophyte water lettuce (*Pistia stratiotes* L.) belongs to the Araceae family (Walsh and Maestro, 2014); the plant spreads predominantly by vegetative propagation (Hussnera et al., 2014) and is native to South America (Hill, 2003). Despite its notorious reputation as an alien plants, water lettuce has also been widely applied in wastewater phytoremediation in tropical areas (Putra et al., 2015) because of its prolific growth characteristics (Chen et al., 2015), great potential in nitrogen and phosphorous removal (Lu et al., 2010), significant absorption, and enrichment in several heavy metals (Lu et al., 2011).

Many studies reported on the use of water hyacinth and water lettuce in phytoremediation to remove different contaminants (Hadad et al., 2011; Lu et al., 2011; Anuradha et al., 2015; Lin and Li, 2016). However, only few comparative studies have explored the purification efficiency of these two macrophytes. In addition, previous studies were mostly conducted under static water conditions; hence, the phytoremediation effects of water hyacinth and water lettuce in sewage treatment cannot be accurately determined under actual liquidity conditions (Sooknah and Wilkie, 2004; Wen et al., 2015). Prior studies reported that removing pollutants from wastewater is inadequate if only one macrophyte is applied in ecological management and suggested plant addition for heavily polluted urban rivers (Hu et al., 2007).

Therefore, the present study aims to determine whether water hyacinth and water lettuce benefit urban sewage phytoremediation in a flowing water system *in situ*, which embodies the fate of nitrogen and phosphorus, particulate matter especially for algal interception, and physicochemical properties of macrophytes. A free-floating, sealed, flow-controlled stainless steel sink was designed as the experimental device for phytoremediation, in which water hyacinth and water lettuce were selected as the test plants. This study may serve as a practical and theoretical reference for contaminant removal from domestic sewage through ecological engineering with water hyacinth or water lettuce.

2. Materials and methods

2.1. Study site

The present study was conducted in a pond (31°17'28.0"N, 119°02'29.3"E; approximately 10500 m² in area, average 2.5 m in depth) located inside the campus of the Jiangsu Academy of Agricultural Sciences, Nanjing, China. The image in the figure was captured using Google Earth on July 13, 2015 (Fig. 1a). The pond water was mainly replenished by domestic sewage and rainwater, and maintained in a eutrophic status with an algal bloom that occurs from May to October every year. Changes of the general physical and chemical parameters of water collected from pond during the experiment were shown as the follows: water temperature (24.9°C–32.2°C), pH (6.98–8.73), DO (4.55 mg L⁻¹–15.73 mg L⁻¹), algae density (3.88 × 10⁷ cells mL⁻¹–15.13 cells mL⁻¹), and chlorophyll *a* concentration (31.81 μg L⁻¹–520.46 μg L⁻¹).

2.2. Experimental device

The cuboid sink was welded with stainless steel (10.0 m × 1.0 m × 0.5 m) and without the top cover, and the sink was maintained in a floating state with bubble float being fixed on both sides of the sink (Fig. 1b, c). Six sinks were placed in experimental area (approximately 200 m²) in this study (Fig. 1d). Water inlet (5 cm in diameter) at one end of the sink was located at 40 cm from the bottom of the sink, and the water outlet at the other end of the sink was controlled by a metering pump

(5 m³ day⁻¹). The metering pump was fixed on the sink and remained at a distance of 20 cm above the water surface. The pumps were semi-enclosed with stainless steel plate to avoid hazards from the rain or burning sun. The metering pump can be running with uniform speed for several months and the pump flow was calibrated at a 24 h interval.

2.3. Experimental plants

The free-floating macrophytes water hyacinth and water lettuce were selected as the test plants. Before the experiment, both plants were cultivated in the “propagation area” fenced with a steel pipe next to the experimental device in the same pond (Fig. 1a). Healthy, similarly sized samples of water hyacinth and water lettuce were collected from the “propagation area” and then respectively placed in two sets of three floating sinks (Nos. 1, 3, and 5 for water hyacinth and Nos. 2, 4, and 6 for water lettuce) with fresh plants of equal weight (7.30 kg m⁻²) (Fig. 1d).

2.4. Water sample collection and analysis

Water samples were obtained from the influent and effluent of all the experimental sinks at a 24 h interval at about 9:00 AM (30 days, from July 7 to August 8 in 2015) for physicochemical parameter analysis. The following parameters were measured *in situ*: water temperature (WT), dissolved oxygen in water (DO), and pH (using a multi-parameter water analyzer, HQ40D, Hach, USA). For water samples used for laboratory analyses, the influent samples (approximately 1 L) were collected from the surface layer (0–0.5 m) at all influent sampling sites, and the effluent samples were obtained from the outlet of the metering pumps (water collected for 1 min, approximately 3.75 L) using 5 L plastic barrels. The physicochemical parameters of the effluent samples were analyzed after being weighed to maintain a constant flow rate daily. Chemical oxygen demand (COD_{Mn}), total phosphorus (TP), orthophosphate (PO₄³⁻), total nitrogen (TN), nitrate (NO₃⁻), ammonium (NH₄⁺), and chlorophyll *a* (Chl *a*) were determined in accordance with standard methods (APHA, 1998). Conversions between COD_{Cr} and COD_{Mn} were as the equation listed below. COD_{Cr} = 6.815 × COD_{Mn} – 1.092 (R² = 0.901), COD_{Mn} (1 mg L⁻¹–10 mg L⁻¹).

For algal species, 0.5 L sub-samples were harvested from 5 L plastic barrels which were collected as described above, and were fixed with 1% acidified Lugol's iodine solution then concentrated to 50 mL by siphon pipe after 48-h sedimentation (Li and Li, 2012). The dominant species was identified with a light microscope at 4000 magnification (CX41, Olympus, Tokyo, Japan). The algal density was determined by using the flow cytometry (FACSJazz, BD company). TN and TP reduce efficiencies of water hyacinth and water lettuce were calculated as the following equation: Reduce efficiency (%) = 100% × (influent concentration – effluent concentration)/influent concentration.

2.5. Plant sample collection and analysis

The biomasses of the water lettuce and water hyacinth were determined and expressed as fresh weight per unit area (kg m⁻²) at a 5 day interval, respectively. Accurate 1 m² plants were harvested from the influent, middle, and effluent sink sites for getting fresh weight with a portable electronic scale. After being weighted, the plants were put back into the sinks neatly (Fig. 1d). In addition, three plants at each site were harvested for laboratory analysis. The shoot and root lengths were measured using a scale ruler before the plants were separated into shoot (stalk and leaf) and root for nutrient analysis.

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