# Potential Role of Halophytic Macrophytes in Saline Effluent Treatment

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Abstract-The growth of the aquaculture industry has been associated with negative environmental impacts through the discharge of raw effluents into the adjacent receiving water bodies. Macrophytes from natural saline lakes, which have adaptability to the high salinity, can be suitable for saline effluent treatment. Eight emergent species from natural saline area were planted in an experimental gravel bed hydroponic mesocosm (GBH) which was treated with effluent water from an intensive fish farm using geothermal water. In order to examine the applicability of the halophytes in treatment processes, we tested the relative efficacy of total nitrogen (TN), total phosphorus (TP), potassium (K), sodium (Na), magnesium (Mg) and calcium (Ca) removal for the saline wastewater treatment. Four of the eight species, which were Phragmites australis, Typha angustifolia, Glyceria maxima, Scirpus lacustris spp. tabernaemontani could survive and contribute the experimental treatment.

*Keywords*—Gravel bed hydroponic system, halophytes, intensive fish farm, salt removal

### I. INTRODUCTION

THE major environmental issue related to the intensive aquaculture production of Hungary is the treatment of nutrient and salt-enriched aquaculture effluents causing physical, chemical and biological changes in the environment. The utilization of saline geothermal water for aquaculture production potentially creates an additional impact if the effluent is higher in salinity than the receiving water bodies.

Constructed wetland treatment systems could provide a simple and low-cost mechanism to treat aquaculture effluents through an integration of physical, biological and chemical

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reactions supported by the significant wetland components [1]-[5]. There are some functional characteristics of macrophytes: plants uptake nutrients directly, provide good conditions for physical filtration reduce water flow and provide a huge surface area for microbial colonisation [6]. Nevertheless a limited efficiency in removing alkali and alkaline cations (e.g., Na, Ca, Mg) in wastewater has been observed in wetland treatment systems [7]-[9]. Associated with primary stresses caused by high salinity, higher plants also suffer from secondary stresses generated by cellular damages [10]. Salinity disrupts the integrity of cell membranes by inducing structural changes and by replacing Ca with Na in the plasma membrane, altering the K/Na ratio [11]. However there are some halophytes from natural saline lakes, which have adaptability to salinity. The protected processes by halophytes should prevent or alleviate the structural and functional damages caused at the cell level. They should also contribute to re-establishment of the homeostatic conditions required for nutrient uptake and intermediate distribution in the presence of an excess of Na<sup>+</sup> and for an internal net flux of water allowing turgor maintenance at the cell level and transpiration at the whole plant level. Halophytes have a number of specific and important mechanisms to achieve crucial protective functions [12]. In the long term, salt accumulation in the plants may impact upon the efficiency of the wetlands in reducing the salt load [13]. Na removal efficiency in wetlands is variable ranging from -78 to 43% [14]-[15]. Sodium and other components of salinity are the most persistent components of recycled water and are among the most difficult removable pollutants from water, usually requiring the use of expensive cation exchange resins or reverse osmosis membranes.

This study was conducted to evaluate the potential role of halophytic macrophytes in purification of specific waste water like saline effluent.

#### II. MATERIALS AND METHODS

The experiments were performed at the Research Institute for Fisheries, Aquaculture and Irrigation (HAKI), Szarvas, in south-eastern Hungary in 2009. The gravel bed hydroponic (GBH) mesocosm were constructed with dimensions of 200 cm wide, 200 cm long and 50 cm deep plastic tanks. Ten centimetres of 1-3 mm gravel layer were filled at the bottom of each unit. One tank was treated with effluent from an intensive African catfish farm and one tank was supplied with river water (Körös) as a control. At both of the tanks, the same

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water flow rate (200 L/day) was applied. The water level was maintained at 0.1 m above the gravel bed surface. Wild plant species, used in the experiment, were collected from a natural saline area located in Homoródszentpál (Sanpaul), mid-eastern Romania. Eight emergent plant species, *Aster tripolium spp. tripolium, Bolboschoenus maritimus, Glyceria maxima, Scirpus lacustris spp. tabernaemontani, Triglochin palustris, Phragmites australis, Typha angustifolia, Carex vulpina,* which are either wetland or salt-tolerant species, were primarily selected for investigations (Table I). These macrophytes were planted in June and after acclimation the tested plots were fed with effluent water and the control plot

 TABLE I

 LIST OF TESTED EMERGENT PLANTS

Scientific name	Common name	Sign
Bolboshoenus maritimus L.	Alkali bulrush	BM
Carex vulpina L.	Great prickly sedge	CV
Glyceria maxima Hartm.	Reed mannagrass	GM
Scirpus lacustris L. ssp. tabernaemontani K.C.Gmel.		
Syme	Soft-stem bulrush	ST
Triglochin palustris L.	Marsh arrowgrass	TP
Aster tripolium L. spp. tripolium	Sea startwort	ATT
Phragmites australis Cav.	Common reed	PA
Typha angustifolia L.	Narrow-leaved cattail	TA

with river water.

The macrophytes were observed throughout the experimental period (90 days) for general appearance and health and were sampled once in 30 days. The fresh and dry biomass of above-ground plant organs (leaves, stems) and below-ground organs (rhizomes, roots) were weighed separately. The plant parts were washed in tap water, rinsed with distilled water to remove metal precipitates or epiphytic microorganisms that might have bound to the surfaces, dried, and ground. To measure the dry weight, the biomass was first dried at 105°C in a drying oven. The powdered samples of the macrophytes were analysed for phosphorus (P), sodium (Na), potassium (K), magnesium (Mg) and calcium (Ca) content using ICP-OES (Application note by Thermo Scientific: 40755), and N was determined by the Kjeldahl method. Water samples were analysed for pH, electrical conductivity (EC), total salt, total suspended solids (TSS), chemical oxygen demand (COD), total nitrogen (TN) and nitrogen forms (NH<sub>4</sub>-N, NO<sub>2</sub>-N, NO<sub>3</sub>-N), total phosphorus (TP) and orthophosphate phosphorus (PO<sub>4</sub><sup>3-</sup>-P) and Na, K, Mg and Ca, chlorid (Cl), sulphate  $(SO_4^{2-})$  and hydrogen-carbonate  $(HCO_3^{-})$  (Table II.) according to the Hungarian Standard Methods (MSZ). Statistical analyses were performed on the growth rate and element accumulation using SPSS software packages. A multiple-range test was used for testing significant differences between the means at the confidence interval of 95%. The data obtained was subjected to an analysis of variance (ANOVA) and the mean differences were compared by LSD tests.

## III. RESULTS AND DISCUSSION

## A. Inlet water quality

The average chemical oxygen demand (COD) of the samples was relatively high (230 mg/L). The total nitrogen content (TN) in the inlet effluent water was about 28.0 mg/L with a maximum peak (35.6 mg/L) measured in June (Table II). The concentration of NO<sub>3</sub>-N ranged between 0.024- $PO_4^{3}-P$ the 0.355 mg/L, and concentration was 1.290±0.335 mg/L. The effluent contained notable concentrations of carbonates (968±75 mg/L), chlorides (28.2±11.4 mg/L) and alkaline metals such as Na, K, Ca, Mg.

 TABLE II

 PARAMETERS OF INLET WATER FOR EXPERIMENTAL WETLAND

 TRFATMENTS

		River water				
Parameters	Unit	Effluent		(control)		
		Average	,	Average		
		(n=8)	s.d.	(n=8)	s.d.	
Conductivity	µS/cm	1357 <sup>a</sup>	54.4	390 <sup>b</sup>	50.4	
$\text{COD}_{\text{Cr}}$	mg/L	230 <sup>a</sup>	34.7	26.9 <sup>b</sup>	19.5	
NH4-N	mg/L	19.7 <sup>a</sup>	4.03	0.130 <sup>b</sup>	0.0745	
NO <sub>2</sub> -N	mg/L	0.016 <sup>a</sup>	0.013	$0.017^{a}$	0.013	
NO <sub>3</sub> <sup>-</sup> -N	mg/L	0.135 <sup>a</sup>	0.108	0.496 <sup>a</sup>	0.341	
TIN	mg/L	18.0 <sup>a</sup>	5.77	0.786 <sup>b</sup>	0.274	
ON	mg/L	8.39 <sup>a</sup>	6.46	1.32 <sup>a</sup>	2.15	
TN	mg/L	28.0 <sup>a</sup>	5.36	2.03 <sup>a</sup>	2.23	
PO <sub>4</sub> <sup>3-</sup> -P	mg/L	1.29 <sup>a</sup>	0.335	0.139 <sup>b</sup>	0.034	
TP	mg/L	3.24 <sup>a</sup>	0.418	0.294 <sup>b</sup>	0.268	
TSS	mg/L	287 <sup>a</sup>	307	28.6 <sup>a</sup>	21.9	
Total salt	mg/L	961 <sup>a</sup>	54.4	270 <sup>b</sup>	59.9	
Ca	mg/L	22.7 <sup>a</sup>	2.31	40.1 <sup>b</sup>	3.65	
K	mg/L	$7.88^{a}$	1.23	4.38 <sup>b</sup>	0.959	
Mg	mg/L	11.6 <sup>a</sup>	0.590	9.11 <sup>b</sup>	1.020	
Na	mg/L	297 <sup>a</sup>	12.1	33.1 <sup>b</sup>	6.5	
Cl	mg/L	28.2 <sup>a</sup>	11.4	31.6 <sup>a</sup>	7.93	
$\mathrm{SO_4}^{2-}$	mg/L	8.15 <sup>a</sup>	3.28	21.8 <sup>b</sup>	10.8	
HCO <sub>3</sub> <sup>-</sup>	mg/L	968 <sup>a</sup>	75.0	181 <sup>b</sup>	16.8	

Values within a row followed by the same letters are not statistically different at p < 0.05 by ANOVA

The average Na content of the samples was significantly higher  $(297\pm12.1 \text{ mg/L})$  than in the control water samples  $(33.1\pm6.5 \text{ mg/L})$ . The Mg, Ca and K concentrations were also significantly higher (p<0.05) than those in the control (Table II.).

## B. Elemental composition of the wild plant species

The initial element concentrations in the plant tissues varied. Ca and Mg accumulated mainly in the roots and Na in the above-ground organs. In the macrophytes, the initial concentrations (in percentage of the dry mass) of Na ranged from 0.547 to 4.42 % in the above-ground organ and 0.456-0.911 % in the below-ground organs, Ca from 0.256 %-1.280 % and Mg from 0,101 %-0.312 % in aerial parts and 0.394-1.115 % and 0.156-0.312 % in the below-ground

tissues, respectively (Fig. 1-Fig. 3). Previous studies [16], [17] reported values of element concentrations in shoots of macrophytes lower than those found in present study. The order of the species in terms of Na content in the biomass was *Triglochin* > *Aster* > *Scirpus* > *Bolboschoenus* > *Carex* > *Typha* > *Phragmites* > *Glyceria* (Fig. 1). With regard to the Ca content, the order of the species was *Typha* > *Carex* > *Aster* > *Scirpus* = *Triglochin* > *Bolboschoenus* > *Glyceria* > *Phragmites* (Fig. 2.). Regarding the Mg levels in the biomass, the order was *Triglochin* > *Typha* > *Carex* > *Aster* > *Scirpus* = *Scirpus* = *Typha* > *Carex* > *Aster* > *Scirpus* = *Triglochin* > *Bolboschoenus* > *Glyceria* > *Phragmites* (Fig. 2.). Regarding the Mg levels in the biomass, the order was *Triglochin* > *Typha* > *Carex* > *Aster* > *Scirpus* > *Bolboschoenus* > *Glyceria* > *Phragmites* (Fig. 3).



Fig. 1 Comparison of the mean above-ground (white) and belowground (shaded) tissue Na concentrations (% in DM) for the eight test species (see Table I. for full species names)



Fig. 2 Comparison of the mean above-ground (white) and belowground (shaded) tissue Ca concentrations (% in DM) for the eight test species (see Table I. for full species names)



Fig. 3 Comparison of the mean above-ground (white) and belowground (shaded) tissue Mg concentrations (% in DM) for the eight test species (see Table I. for full species names)

#### C. Plant growth and nutrient uptake

Four out of the eight species, which were Bolboschoenus (BM), Carex (CV), Triglochin (TP) and Aster (ATT) showed unhealthy symptoms from the combined effect of high salt concentration and flood conditions. They appeared dry and yellowish and retardation was observed. Oxygen restriction, which is resulted by prolonged flood conditions as well as organic waste, caused the growth of these plants to suffer. The other species, which were Typha (TA), Phragmites (PA), Glyceria (GM) and Scirpus (ST), exhibited a stress tolerance and also may have a potential role in wastewater treatment. After 90 days growth, Typha was more productive (aerial biomass of 31.2 kg/m<sup>2</sup>) than the others like Scirpus  $(10.98 \text{ kg/m}^2)$ , Glyceria  $(8.68 \text{ kg/m}^2)$  and Phragmites  $(2.24 \text{ kg/m}^2)$  in wet mass. The biomass values of the control tank were lower, the aerial organs of Typha:  $0.886 \text{ kg/m}^2$ , the Scirpus: 0.261 kg/m<sup>2</sup>, the Phragmites: 0.611 kg/m<sup>2</sup>, and the Glyceria:  $0,700 \text{ kg/m}^2$ .

The element uptake (i.e. the quantity of element removed by the plant) was influenced by the plant issue concentration and repartition of the organs in the harvested biomass. "Fig 4-Fig 9"-show the nutrient uptake in the biomass of the tested plants. The nutrient uptake rates of the aerial biomass were in the range of 0.218-1.38 g/m<sup>2</sup>/d, 0.019-0.176 g/m<sup>2</sup>/d and 0.154-0.602 g/m<sup>2</sup>/d for N, P and K, respectively. Tanner [16] reported N uptake rates of eight macrophytes similar to those found in present study, the mean uptake rates were 0.744±0.072 g/m<sup>2</sup>/d. and observed lower uptake rates of P  $(0.104\pm0.007 \text{ g/m}^2/\text{d})$ . The rate of K  $(1.03\pm0.065 \text{ g/m}^2/\text{d})$  was higher compared to the levels found in biomass in the present study. The N uptake rates differed among the tested species at p<0.05 and Typha showed the highest rate. In comparison, Klomjek [18] reported a lower N uptake rate of 0.061 g/m<sup>2</sup>/d and P uptake rate of 0.00024 g/m<sup>2</sup>/d at Typha species in a treatment wetland receiving a lower N loading rate.



Fig. 4 Uptake  $(g/m^2/day)$  of N and repartition among the plant organs, mean above-ground (white) and below-ground (shaded) and control (grey) (see Table I. for full species names)

A similar trend was observed for the P uptake rate, *Typha* was the most efficient plant in this category. Nonetheless, the P uptake rate showed difference among the different species, *Phragmites* and *Glyceria* at p<0.05. A higher K accumulation was observed in the aerial biomass, without significant differences between the species (p<0.05).



Fig. 5 Uptake  $(g/m^2/day)$  P and repartition among the plant organs, mean above-ground (white) and below-ground (shaded) and control (grey) (see Table I. for full species names)



Fig. 6 Uptake  $(g/m^2/day)$  of K and repartition among the plant organs, mean above-ground (white) and below-ground (shaded) and control (grey), (see Table I. for full species names)

The nutrient uptake rates were in the range of 0.036-1.449 g/m<sup>2</sup>/d, 0.022-0.426 g/m<sup>2</sup>/d and 0.022-0.156 g/m<sup>2</sup>/d for Na, Ca and Mg, respectively. The Na uptake rate differed among the tested species at p<0.05 and *Phragmites* showed the lowest rate (0.036 g/m<sup>2</sup>/d), its Na uptake rate statistically differed from that of *Glyceria* (0.218 g/m<sup>2</sup>/d), *Scirpus* (0.522 g/m<sup>2</sup>/d) and *Typha* (1.44 g/m<sup>2</sup>/d) p<0.05 in aerial biomass.



Fig. 7 Uptake  $(g/m^2/day)$  of Na and repartition among the plant organs, mean above-ground (white) and below-ground (shaded) and control (grey) (see Table I. for full species names)



Fig. 8 Uptake  $(g/m^2/day)$  of Ca and repartition among the plant organs, mean above-ground (white) and below-ground (shaded) and control (grey) (see Table I. for full species names)



Fig. 9 Uptake (g/m<sup>2</sup>/day) of Mg and repartition among the plant

organs, mean above-ground (white) and below-ground (shaded) and control (grey), (see Table I. for full species names)

Ca and Mg were accumulated mainly in the above-ground organs at significantly higher rate in *Typha* than the other species, with values of  $0.426 \text{ g/m}^2/\text{d}$  and  $0.156 \text{ g/m}^2/\text{d}$ , respectively. The Ca uptake rate of *Typha* statistically differed from *Glyceria*, *Scirpus* and *Phragmites* at p<0.05. Harvesting the above-ground biomass is therefore a possibility to remove accumulated elements.

#### IV. CONCLUSION

An understanding of the importance and sustainability of different nutrient removal processes is necessary to improve the longer-term capabilities of constructed wetland systems [19]. The feasibility of using salt-tolerant plants (halophytes) as biofilters may remove nutrients and salt components from the saline aquaculture effluents. *Typha, Phragmites, Glyceria* and *Scirpus* could survive and facilitate the experimental treatment, so the salt-tolerant plant species may provide a suitable alternative for constructed wetlands receiving effluent water loaded by salinity and plant nutrient.

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