Revista CIENCIA **Vol. 20, 1, 73-83 (2018)**
http://dx.doi.org/10.24133/ciencia.v20i1.557

"Typha latifolia" effective phosphorous extractor from hydromorphic soils: comparison between populations local growing and Technosol eutrophic

A.I. Simões [(1)]; X. L. Otero, [(*1)]; F. Macías [(1)]; P. Guevara [(1,2)]

[1] Dep. Edafología y Química Agrícola. Facultad de Biología. Universidad de Santiago de Compostela,
Campus Sur. 15782, Spain. : xl.otero@usc.es
[2]Dep. Ciencias de la Tierra y la Construcción. Universidad de las Fuerzas Armadas
ESPE. Sangolqui-Ecuador. pvguevara@espe.edu.ec
*Corresponding author: e-mail: xl.otero@usc.es;

Received: Agosto 2017. Accepted: December 2017

ABSTRACT

Eutrophication of soils and waters is one of the main problems that threaten the conservation of species and habitats. Recent studies show that P can also substantially affect the loss of biodiversity. In contrast to N, which can be removed by leaching, P remains over time because it is adsorbed to soil colloids. A study to evaluate the common cattail (Typha latifolia) ability to extract phosphorous from a eutrophic Technosol (ET), highly enriched with P, and from an oligotrophic natural soil (NS) was performed. In the soils the total content of nutrients (P, Ca, Mg, K, N) Fe, Al, C, amorphous oxyhydroxides of Al/Fe, bioavailability P and geochemical forms of P was performed. The results showed a significantly high concentration of the TP and bioavailability P in ET than NS. In line with a high concentration of P in ET, a very high concentration of P was extracted by the plants in ET. These results showed that cattail responds positively to very high concentrations of P in the soil and, therefore, must be considered as a very efficient species to reduce soil eutrophication.

Keywords: Wastewaters, Phosphorous, Eutrophication, *Typha latifolia* Eutrophic Technosol

"Typha latifolia" extractor eficaz de fósforo en suelos hidromórficos: comparación entre poblaciones de cultivo local y Tecnosol eutrófico

RESUMEN

La eutrofización de suelos y aguas es uno de los principales problemas que amenazan la conservación de especies y hábitats. Estudios recientes muestran que el P también puede afectar sustancialmente a la pérdida de biodiversidad. En contraste con el N, que puede eliminarse por lixiviación, el P es adsorbido por el suelo pudiendo permanecer a lo largo del tiempo. Se realizó un estudio para evaluar la capacidad de la Typha latifolia para extraer el P del suelo. Para ello se consideró un Technosol eutrófico (ET), altamente enriquecido con P y un suelo natural oligotrófico (NS). En los suelos se analizó el contenido total de Fe, Al, C orgánico, oxihidróxidos amorfos de Al/Fe, formas geoquímicas de P. Los resultados muestran una concentración significativamente más elevada de P total y biodisponible en ET. En consonancia con una alta concentración de P en ET, la población de Thypha mostró también una concentración significativa superior de P comparando con las que crecieron en el NS. Estos resultados muestran que la totora responde positivamente a concentraciones muy elevadas de P en el medio y, por lo tanto, debe ser considerada como una especie muy eficiente para reducir la eutrofización del suelo.

Palabras clave: Aguas residuales, Fósforo, Eutrofización, Typha latifolia Technosol eutrófico.

1. INTRODUCTION

Discharge of wastewater from agricultural and industrial activities contributes to P enrichment in aquatic ecosystems (Camargo et al., 2005) leading to eutrophication. Eutrophication can be defined as the enrichment of waters by nutrients and the consequent deterioration of quality due to the luxuriant growth of plant life, and its repercussions on the ecological balance of the waters affected (Yeoman et al., 1988). The limiting nutrient in most freshwater lakes, reservoirs and rivers is phosphorus, and inputs of this element from anthropogenic sources accelerate the process of eutrophication, which normally proceeds slowly in the natural ageing of lakes (Pastor et al., 2008).

Traditionally, macrophytes have been suggested for the treatment of industrial or sanitary effluents in constructed wetlands (CWs) (Campos, 1999; Ferreira et al., 2003; Mannarino et al., 2006). Through the mechanisms of absorption and assimilation, the plants are able to remove nutrients from polluted waters, many of which are trapped in the system by successive cycles of growth, death and decomposition (Kadlec et al., 2005).

The concentration of ions, such as phosphates, in liquid effluents generated in wastewater treatment may be low, in relation to the demand of the macrophytes grown in this medium. As these ions are essential for the production of biomass and, consequently, for the development of macrophytes, the efficiency of these plants in the effluents treatment is directly related to the capacity of absorption and extraction of nutrients (Campos, 1999). This occurs even though the content of these ions in these liquids is generally greater than the maximum limits allowed by the environmental bodies for the emission of effluents into watercourses. In general, the nutrient extraction capacity of macrophytes increases with the growth of these plants (Roston and Mansor, 1999).

Typha latifolia stands out among the species of macrophytes most used in CWs for wastewater treatment. This species is adapted to flooded soils and sediments and produces large amounts of biomass (Ferreira et al., 2003; Mannarino et al., 2006).The efficiency of the macrophytes in removing P varies according to the type of effluent. A study carried out with domestic sewage showed greater removal of orthophosphate with Typha latifolia than with Eleocharis fistulosa (Mansor, 1998). When it was cultivated in bed with gravel and wastewater from anaerobic reactor, the efficiency of Typha latifolia in removing P from this effluent was 25% (Mazzola, 2005). Although this efficiency is considered low, it was higher than Eleocharis fistulosa, which coincides with that observed by Roston and Mansor (1999). For Typha latifolia and other macrophytes, both aspects of growth and nutrition have been investigated in numerous studies (Boyd, 1970). Nevertheless, most of the plants have been developed in natural wetlands with a low level of nutrients and there are very few data referred to rich nutrient or polluted areas, since the research carried out in wastewaters has been mainly focused on the study of the water quality rather than the development of the plants and their ability to extract nutrients.

To clarify Typha latifolia's efficiency to extract phosphorus from eutrophic soils highly enriched with phosphorous, this study was carried out. For that, it is compared P extraction efficiency of a Typha latifolia population grown in a eutrophic Technosol highly enriched with phosphorous (ET) in a constructed wetland (CW) and the efficiency of extraction of a local population grown on natural soil (NS).

2. MATERIALS AND METHODS

2.1 Field sampling and initial soil characterization

Soils and plants were collected from an area of Nature 2000 Network (natural area) (NO Spain; 42°32'51.8"N 8°22'34.2"W) and from a constructed wetland (CW) (NO Spain; 42°32'55.7"N 8°42'36.7"W) designed to reduce concentration of dissolved organic matter from discharges from a winery. Both are located in Pontevedra province (Galicia, NW Spain). Natural soil (NS) and eutrophic Technosol (ET) composite soil samples (mixture of three simple samples) from soil surface (top 20 cm) coming from natural area and from CW were collected. Twelve plants of *Typha latifolia* (AT) were sampling from ET and four from the control area (NS) so-called NT.

In spring period, AT and NT plants were cut at ground level and all aerial biomass was collected and kept in plastic bags until arrival at the laboratory. The best conserved parts of the plant were selected, eliminating the dry or damaged ones. The aerial biomass (stems and leaves) was cut into small pieces. ET and NS composite soil samples (mixture of three simple samples) from 0- to 20- cm layer were collected at each of the three sampling points distributed in zigzag in each established sampling unit.

2.2 Plant analysis

Plant material was carefully washed with deionized water and dried at 45ºC until constant weight. The dry weight of leaves and stems corresponded with the total aerial biomass. Plants TP content was measured in an UV-VIS spectrophotometer (Perkin Elmer-1100B model) following digestion with concentrated nitric acid and following vanado-molibdo-phosphoric acid colorimetric method (Jackson, 1970)

2.3 *Soil analysis*

One part of the soil samples were dried at 45ºC until constant weight and sieved (2mm) before analytical determinations. The pH and Eh were measured in situ using specific electrodes. Particle size distribution was determined by

Robinson's pipette method, following organic matter oxidation with H2O2 and dispersion with $Na_6P_6O_{18}$. Total P, Ca, K, Na, Mg, Fe, Al and Mn concentrations were measured after digestion of soil samples in a nitric acid - perchloric acid mixture (Olsen and Sommers 1982). TP concentration and in the extractant following vanado-molibdo-phosphoric acid colorimetric method (Jackson, 1970). C and N contents were determined by total combustion in an elemental analysis apparatus Leco Truspec CHN. Amorphous oxyhydroxides of Fe (Fe-OX) and Al (Al-OX) were extracted with acid ammonium oxalate (Blakemore 1977). The concentration Ca, K, Na, Mg, Fe, Al and Mn were measured by AAS in a Perkin Elmer-1100B apparatus.

Partitioning of P was determined in wet soil samlples (1 g), using a sequential extraction method that enables identification of seven operationally defined P fractions, which are described as follows (for further details, see Ruttenberg, 1992; Otero et al., 2015).

F1: (ads-P), weakly adsorbed and soluble phosphorus. The samples were extracted with 20 ml of 1 M MgCl2 solution, with continuous stirring for 30 m. F2 (Fe/Mn-P), P adsorbed to Fe oxides and oxyhydroxides. 20 ml of a sodium bicarbonate-dithionite solution (Na2S2O4 0.11 M + 0.11 M NaHCO3; pH 7) was added to the residue from the prior extraction, and the mixture was stirred continuously for 1 h. F3 (Al-P), P bound to Al hydroxides and clay. The samples were shaken for 18 h with 20 ml of sodium hydroxide (0.1 M NaOH). F4 (HA-P) P associated with soil humic substances. the extract was acidified with 2.5 ml of concentrated sulphuric acid (H2SO4) and allowed to stand overnight (Hedley et al., 1982)., were then separated by filtration. The filter was dried (45 °C) and calcined at 520 °C for 2 h in a muffle furnace. The ashes were dissolved in 5 ml of 1 M HCl by boiling the mixture for 30 min, and the extract was made up to 50 ml with Milli-Q water., F5 (Ca-P), P bound to Ca phosphates (i.e. apatite). The samples were shaken for 1 h with 20 ml of 0.5 M HCl. F6 (Res-P) P mainly associated with refractory organic matter (Schlichting et al., 2002), although some mineral-bound P not extracted with alkali or acid can be included (Reddy et al., 1999). The material remaining after the above extractions was calcined and processed as described in step F4(HA-P).

2.4. Statistical Analysis

Statistical analysis was performed with SigmaStat 3.5 07; differences were considered significant at $p < 0.05$. For aerial plant tissue (stems and leaves), differences in total dry biomass and total phosphorus (TP) concentration were statistically compared by Mean-Whitney Rank Sum Test. Differences in TP, Calcium (Ca), iron (Fe), aluminium (Al), total nitrogen (TN), total carbon (TC), aluminium and Iron oxides and hydroxides (Al-OX and Fe-OX) concentrations in both soils were statistically compared by Mean-Whitney Rank Sum Test.

3 RESULTS AND DISCUSSION

3.1 General characterization of ET and NS soils

The ET and NS soils showed the same texture class but different conditions Eh-pH (Table 1). Soil pH was acid in NS and neutral in ET, whereas Eh was oxic in ET and suboxic in NS (Brooking, 1982). Soil pH and redox status (Eh) strongly influence the phosphorus solubility in wetland soils. Thus, at oxic conditions (Eh > 350 mV) phosphorus solubility is low, resulting in low phosphorus concentration in soil solution whereas at sub-oxic and anoxic condition (Eh < 300 mV), phosphorus solubility increases at all pH values, reflecting high concentration in soil pore water (Patrick and Khalid, 1974). In this sense, the P is thought to be more bioavailable in NS than in ET.

Table 1. Main soils components and properties

Parameter	ET Soil	NS Soil
Ph	7,2	5,6
Eh (mV)	455	256
TP (mg kg^{-1})	11841	1601
Ca (mg kg^{-1})	67960	1880
K (mg kg^{-1})	6101	6030
Na (mg kg^{-1})	835	147
Mg (mg kg^{-1})	8480	2800
Fe (mg kg^{-1})	18244	17880
Al (mg kg^{-1})	25146	16780
Mn (mg kg^{-1})	2502	294
Al-Oxa*(* mg kg^{-1})	15851	4390
Fe-Oxa(mg kg^{-1})	6192	3933
TC (%)	11,26	3,69
TN (%)	0,77	0,26
Texture	Sandy loam	Sandy loam

Concentration of TC, TN (Figure. 1 A), Ca, Fe and Al (Figure. 1 B) were significantly higher in ET than NS soil (*p* < 0,001) (TC: ET = 11,2 % , NS = 3,7 %; TN: ET= 0,77 mg kg $^{-1}$,NS = 0,26mg kg $^{-1}$; Ca: ET = 67960 mg kg $^{-1}$,NS = 1880 mg kg $^{-1}$; Fe: ET = 18244 mg kg $^{-1}$,NS = 17880 mg kg $^{-1}$; Al: ET = 25146 mg kg $^{-1}$,NS = 16780 mg kg $^{-1}$). The higher content of Ca is consonant with the incorporation of large amounts of calcium carbonate in ET soil. Additionally, the high content of Al in ET soil is explained by the incorporation of aluminium hydroxide during Technosol elaboration. ET soils showed also a higher content of total carbon (TC ~11% vs. ~4%) and nitrogen (TN ~0.8% vs. ~0.3%) than NS. This high incorporation of nutrients in ET soils was performed to get a fast macrophytes development to reduce dissolved organic matter produced by winery.

The statistical comparison between ET and NS soil relate to Al-OX and Fe-OX contents is shown in Figure. 1 C. It was observed a significantly higher concentration (*p* < 0,001) of Al-OX and Fe-OX at ET soil than at NS soil (Al-OX: ET= 15851 mg kg $^{-1}$, NS= 4390 mg kg $^{-1}$; Fe-OX: ET=6192mg kg^{-1}, NS= 3933 mg kg^{-1}). The higher values of Al-OX and Fe-OX in ET soil are in agreement with the incorporation of high amounts of amorphous-Al/Fe oxyhydroxides in ET.

Several researchers reported significant correlations between amorphous and poorly crystalline forms of iron (Fe-OX) and aluminium (Al-OX), with phosphorus retention of soils (Khalid et al., 1977). Phosphate associated with Fe and Al oxyhydroxides are stable under acidic soil conditions, and P solubility is higher as pH increases (Patrick and Khalid, 1974).

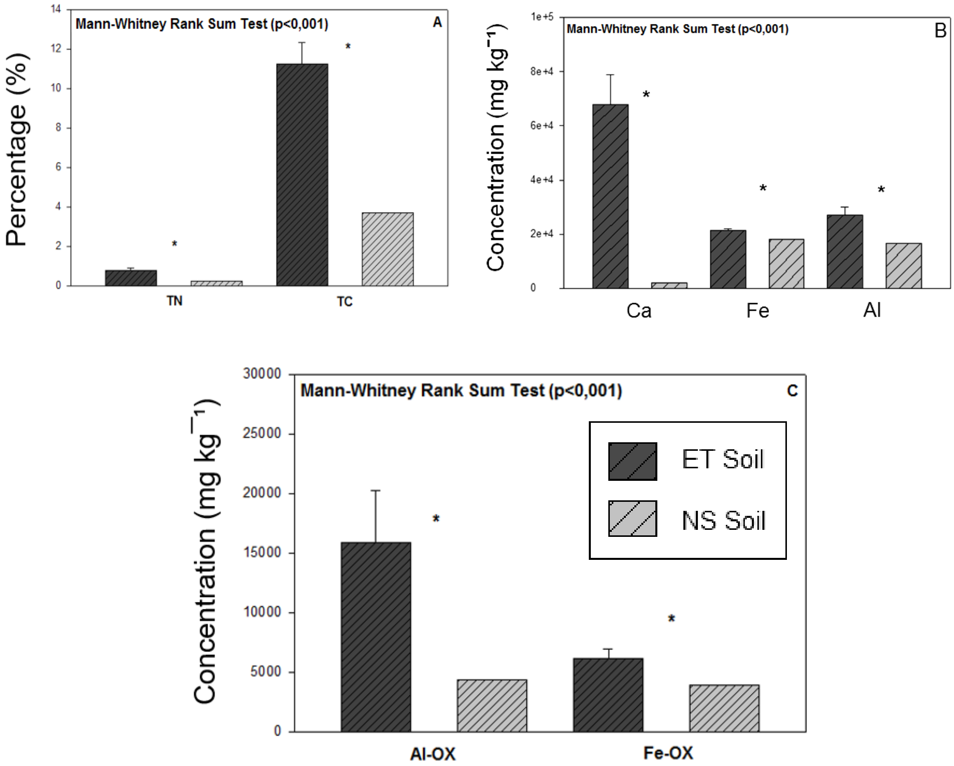

Figure. 1. A) Concentration of total carbon (TC) and total nitrogen (TN) in eutrophic Technosol (ET) and natural soil (NS). **B)** Concentration of calcium (Ca), iron (Fe) and aluminium (Al) in ET and NS soils. **C)** Concentration of amorphous aluminium (Al-OX) and amorphous iron (Fe-OX) in ET and NS soils at spring period. Values are average and bars on columns the standard error (n=3). (*) indicate significant differences between ET and NS soils (Mann-Whitney Rank Sum Test, $p < 0,001$).

3.2. Total (TP) and partitioning of P in ET soil

In order to realize the effect of a greater content of phosphorus in soil and the contribution of soluble P fractions for plant growth and, consequently, for P extraction, it were evaluated the differences of TP content between ET and NS soils and the different P fractions of the ET soil obtained by partitioning.

The statistical comparison of TP concentration is shown in Figure. 2 A. It was observed a significantly higher ($p < 0,001$) concentration of TP in ET soil (11841mg kg^{-1}) compared to NS soil (1601 mg kg^{-1}). This is in agreement with the incorporation of phosphate fertilizers during the ET elaboration

Regarding to P partitioning in ET soil, Ca-P was the most representative P fraction in this soil and showed a concentration of 7383 mg kg-1. This great quantity is allusive to the incorporation of large amounts of calcium phosphate in this soil. The Clay/Al-P fraction showed a concentration of 3356 mg kg-1 and it was the second most representative P fractions in ET soil. This value is related to the incorporation of a great quantity of hydroxides of Al in this soil. The Fe/Mn-P fraction concentration was 438 mg kg-1. This value is consonant with the incorporation of considerable amounts of oxides and hydroxides of Fe and Mn in ET soil. The Res-P fraction had a concentration of 593 mg kg-1 and it is characterized as a recalcitrant phosphorus fraction, which do not contribute to plant nutrition, except in extreme soil deficiency (Gatiboni et al., 2007). Relate to the HA-P fraction, it showed a concentration of 317 mg kg-1. This high value may be due to the state of development of the plants in the spring period, which allow a strong production of root and microbial exudates that are important sources of humic acids in the soil

Finally, the concentration of Ads-P, considered to be the soluble soil P and plant available (Foth and Ellis, 1997), was the less representative P fraction in ET soil, although showed a concentration of 111 mg kg-1 (Fig. 2), which is a very high value when compared to that obtained in granitic acid soils from Galicia with a concentration of Ads-P < 15 mg kg-1 (Macías et al., 1982), due to high concentration of amorphous oxy-hydroxides of Fe/Al, which strongly adsorb or precipitate the phosphate ion P (Foth and Ellis, 1997). This fraction represents the set of phosphate compounds capable of rapidly replenishing the soil solution when it is absorbed by plants or microorganisms (Walker and Syers, 1976)

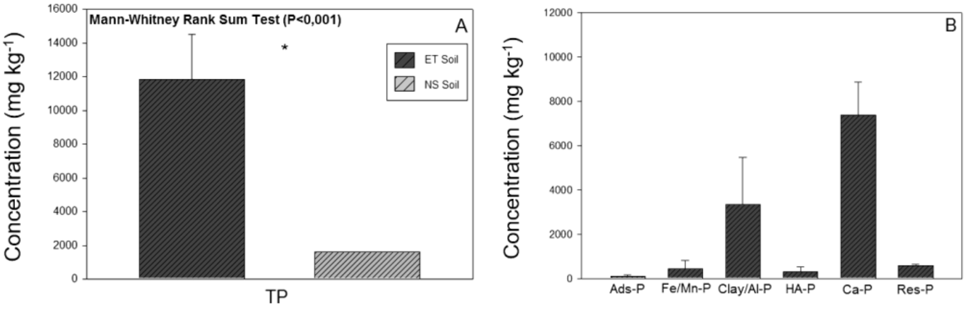

Figure. 2. A) Total P (TP) in eutrophic Technosol (ET) and natural soil (NS). Values are average and bars on columns the standard error (n=3). (*) indicate significant differences for the TP concentrations between ET and NS soils (Mann-Whitney Rank Sum Test, *p* < 0,001). **B)** P fractions in ET soil at spring period. Values are average and bars on columns the standard error (n=3)

P is an important macronutrient, being a key component of molecules such nucleic acids, phospholipids, and ATP, and, consequently, plants cannot grow without a reliable supply of this nutrient. After N, P is the second most frequently limiting macronutrient for plant growth (Foth and Ellis, 1997).

Having into account that the concentration of this nutrient in soil solution is generally low and plant requirements are high, the greater content of TP in ET soil may have contributed to the plant growth enhancement. Our results also shown the capacity of ET soil to provide the most soluble P (ads-P) at greater concentrations to plants, when compared to supply ability of natural Galician soils.

3.3. *Typha latifolia aerial biomass and phosphorus (P) accumulation in plants*

No significant differences were found in *T. latifolia* aerial biomass between AT and NT population, although a tendency to show lower biomass of leaves and stems was observed in NT plants (Aerial biomass: ET=57 g d.w., NT= 23 g d.w.) (Figure. 3 A).

The aerial productivity of AT plants was estimated as 2451 g m^{-2}. This productivity was higher than that obtained by NT plants (989 g m^{-2}) and higher than the range of 430-1480 g m^{-2} obtained for Typha spp. above biomass in natural stands (Dubbe et al., 1989). Thus, the higher response of T. latifolia to the great quantity of nutrients in ET soil is clear.

Relate to TP concentration extracted by plants, AT population showed a significantly higher concentration ($p < 0,001$), when compared to NT plants (TP: AT= 8960 mg P kg^{-1} d.w., NT= 3507 mg PKg^{-1} d.w.) (Figure. 3 B). The total phosphorus extraction carried out by the aerial part of AT plants grown on ET soil was estimated as 12,88 kg P $year^{-1}$.This extraction was significantly higher than that obtained for NT (2,03 Kg P $year^{-1}$) and in cattail stands in non-polluted areas, whose productivities are much lower (Dubbe et al., 1989).

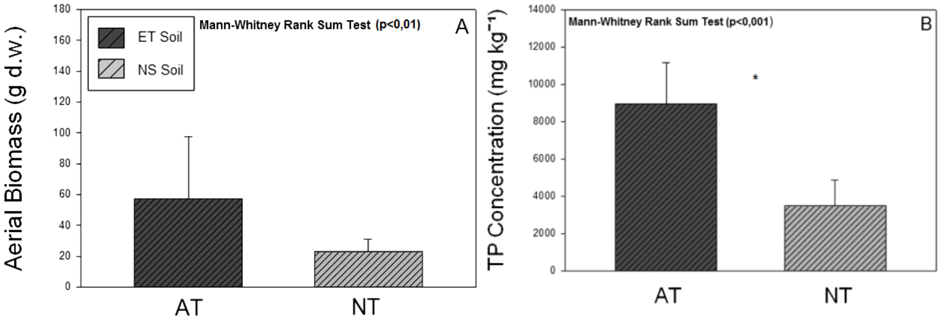

Figure 3 A) Aerial Biomass and **B)** total phosphorus in *T. latifolia* growing in the ET (AT) and NS areas. Values are average and the bars on columns the standard error (n=3). ET (eutrophic Technosol highly enriched with phosphorus). NS (natural soil). (*) indicate significant differences between ET and NS soils (Mann-Whitney Rank Sum Test, p<0,001).

Many studies have pointed the ability of macrophytes to remove P and pollutants, both in natural marshes (Lee et al., 1975; Kadlec and Tilton, 1979) and artificial systems (Jong, 1976; Kickuth, 1984).

Typha latifolia have been known to be a particularly effective substrate in removing nutrients from wastewater because of relatively high biomass above and below ground, providing potentially greater surface area for the uptake of nutrients and ions (Shutes, 2001). It have been stated that the role of plants in P extracting was also influenced by factors such as nutrient loading rates, the season of the year and the physiological state of plants (Zhao et al., 2010). This means that large amounts of biomass produced by plants results in considerable nutrient extraction during the growth period (Dubbe et al., 1989).

Put this, we conclude that the higher growth rate of the aerial biomass observed for AT specimens contributed to the greater accumulation of TP. Our results also confirmed the hypothesis that *T. latifolia* is an efficient species for extraction of P from natural environment and that extracts significantly more P in eutrophic environments than in undisturbed ones, although the development of the aerial part of the plant did not present significant differences between the two environments. However, the same trend was observed between the extraction of P carried out by the plants and the accumulation of biomass. It was also proved that *T. latifolia* is a very efficient species in extracting P also from more complex matrices such as the Technosols. In this sense, it is worth noting that Technosols (WRB, 2007) are a new unit of soils, that can be technically designed (Technosol "à la carte"), to strengthen the attenuation capacity of natural systems, correct dysfunctions and increase the damping capacity and the critical load of pollutants in the soil-water system. (Macías-García, 2006; Macías, 2007).

4. CONCLUSIONS AND PRACTICAL CONSIDERATIONS

The present study showed that *T. latifolia* is an efficient species in reducing P from eutrophic soil. *Typha latifolia* growing in a eutrofic Technosol highly enriched with phosphorus, was able to remove amounts significantly higher than the control area.

From the point of view of wastewater depuration, the construction of CWs should consider the most suitable macrophyte species for that purpose, favoring the choice of native species to reduce the risk of invasion of exotic species, mainly in protected areas. In addition, prior to the election of the species, consideration should be given to the composition and properties of the soil to choose the species that may have a better and faster development and that allows greater efficiency in the removal of the contaminants.

5. ACKNOWLEDGEMENTS

Support for this research was provided by the Xunta de Galicia (Spain). The authors would like to thank to the staff of “Pazo de Señorans” winery for their collaboration and allowance to collecting the samples.

REFERENCES

1. Camargo, M. S. et al., (2005). “Extractors of silicon soluble in soils: influence of limestone and phosphorus”. *Biosci. Journal, Uberlândia*. **2** , 9-19.
2. Yeoman, S., Stephenson, T., Lester, J.N. and Parry, R., (1988). “The removal of phosphorus during wastewater treatment: A review”. *Environmental Pollution* **49**, 183-223.

3. Pastor L., Marti N., Bouzas A., Seco A., (2008). “Sewage sludge management for phosphorus recovery as struvite in EBPR wastewater treatment plants, Bioresour”. *Technol*. **99**, 4817–4824.
4. Campos, J.R., cood., (1999). “Tratamento de esgotos sanitários por processo anaeróbio e disposição controlada no solo”. São Carlos, PROSABE-FINEP. 435.
5. Ferreira, J.A.; Campos, J.C.; Ritter, E. and Mannarino, C.F., (2003). Wetland: Resultados no tratamento do chorume do aterro sanitário de Piraí-RJ. In: Congresso Brasileiro de Engenharia Sanitária e Aambiental, **22**., Rio de Janeiro, 2003. Anais. Joinville, Associação Brasileira de Engenharia Sanitária e Ambiental 1-10.
6. Mannarino, C.F.; Ferreira, J.A.; Campos, J.C. & Ritter, E., (2006). “Wetland para tratamento de lixiviados de aterros sanitários – experiências no aterro sanitário de Piraí e no aterro metropolitano de Gramacho”. *Eng. Sanitária Amb.* **11,** 108-112.
7. Kadlec, R.H., (2005). “Phosphorus Removal in Emergent Free Surface Wetlands”. *J. Environ. Sci. Health A.* **40,** 6–7, 1293– 1306.
8. Roston, D.M. and Mansor, M.T.C., (1999). Sewage treatment by a system of cultivated beds of sub-superficial flow: Evaluation of nitrogen removal. In: Brazilian Congress of Enrionmental Sanitary Engineering, **20.** Anais, Rio de Janeiro, Braz. Assoc. Environ. Sanit. Engineering. 519-524.
9. Mansor, M.T.C., (1998). “*Uso de leito de macrófitas no tratamento de águas residuárias*”. Campinas, Universidade Estadual de Campinas. (Tese de Mestrado) 106.
10. Mazzola, M., (2005). “Uso de leitos cultivados de fluxo vertical por batelada no pós-tratamento de efluente de reator anaeróbio compartimentado”. *R. Bras. Eng. Agríc. Amb*. **9**, 276-283.
11. Boyd, C.E., (1970). “Production, Mineral Accumulation and Pigment Concentrations in Typha Latifolia and Scirpus Americanus”. *Ecology* vol. **51** (2), 285-290.
12. Jackson, M.L.,. (1970). “*Análisis Químico de Suelos*”. 2ª ed. MARTINEZ, J.B., Trad. Barcelona, Ed. Omega S.A. 662.
13. Ruttenberg, K.C., (1992). “Development of a sequential extraction method for different forms of phosphorus in marine sediments”. Limnol. *Oceanogr***.** **37**(7), 1460-1482.
14. Otero, M., Santos, D., Barros, A.C., Calapez, P., Maia, P., Keizer, J.J., Esteves, V.I., Lillebø, A.I., (2015). “Soil properties, phosphorus fractions and sorption after wildfire in northcentral Portugal”. *Geoderma Reg*. **5**, 86–95.
15. Schlichting, A. Leinweber, P., Meissner, R., Altermann, M., (2002). “Sequentally extracted phosphorus fractions in peat-derived soils”. *J. Plant Nutr. Soil Sci.* 165, 290-298.
16. Reddy, D.D., Rao, A.S., Takkar , P.N., (1999). “Effects of repeated manure and fertilizer phosphorus additions on soil phosphorus dynamics under a soybean-wheat rotation”. *Biol. Fertil. Soils*. **28**, 150-155.
17. Brooking,(1982). “Eh-pH diagrams for geochemistty”. *Ed. Springer*.
18. Patrick, W.H., Jr., and R.A. Khalid, (1974). “Phosphate release and sorption by soils and sediments: Effects of aerobic and anaerobic conditions”. *Science* **186**, 53-55.

19. Gatiboni, L.C.; Kaminski, J.; Rheinheimer, D. dos S.; Kaminski, J.; Flores, J.P.C., (2007). "Biodisponibilidade de formas de fósforo acumuladas em solo sob sistema plantio direto". *Rev. Brasi. Ciênc. Solo* **31**, 691-699.
20. Foth, H.D. and Ellis, B.G., (1997). "*Soil fertility*". 2.ed. Boca Raton: Lewis Publishers. 304.
21. Walker, T. and Syers, J., (1976) : "The fate of phosphorus during pedogenesis", *Geoderma* **15**, 1–19.
22. Dubbe D.R., Garver E. G. and Pratt D. C, (1989). "Production of Cattail (Typha spp.) biomass in Minnesota, USA". *Biomass* **17**, 79-104.
23. Lee, G.F., Sonzogni W. and Spear R.D., (1977). *Significance of oxic versus anoxic conditions for Lake Mendota phosphorus release. In: Interactions between sediments and fresh water*. Golterman, H. (ed.) Springier-Verlag, New York, NY. 294-306.
24. Jong, J., (1976). *The purification of wastewater with the aid of rush or reed ponds*. In J. Tourbier and K.W. Pierson, Jr. (eds.) Biological Control of Water Pollution. University of Pennsylvania Press, Philadelphia. 133-139.
25. Kadlec, R.H. and Tilton, D.L., (1979). "The use of freshwater wetlands as a tertiary wastewater treatment alternative". *CRC Critical Reviews in Environ. Contr.* **9**, 185-212.
26. Kickuth, R., (1984). "Das Wurzelraumverfahren in der Praxis". In Landschaft und Stadt.**16**, 145-153.
27. Shutes, R.B.E., (2001). Artificial wetlands and water quality improvement. *Environ. Internat*. **26**, 441-447.
28. Zhao, Y.J., Liu, B., Zhang, W.G., Ouyang, Y., Qing An, S., (2010). "Performance of pilotscale vertical-flow constructed wetlands in responding to variation in influent C/N ratios of simulated urban sewage". *Bioresour. Technol.* **101**, 1693, 1700.
29. WRB. FAO, (2007). *World reference base for soil resources*. First atualization 2007. World Soil Resources Reports No. 103. FAO, Rome.
30. Macías-García, F., (2006). *Elaboración de Suelos derivados de residuos con diferentes aplicaciones ambientales.* DEA. Medio Ambiente y Recursos Naturales. Universidad de Santiago de Compostela. Santiago de Compostela: s.n.
31. Macías, F.; Calvo de Anta, R; García, C.; Garcia-Rodeja, E; Silva, B.; (1982). "El material original. Su formación e influencia en las propiedades de los suelos de Galicia". *An. Edaf. Agrob*. **41**, 1747-1768.
32. Hedley, M. J., J. W. B. Stewart, and B. S. Chauhan, (1982). "Changes in inorganic and organic soil phosphorus fractions by cultivation practices and by laboratory incubations*". Soil Sci. Soc. Am. J*. **46,** 970-976.
33. Macías, F., Bao, M., Macías-García, F. & Camps, M., (2007)." Valorización biogeoquímica de residuos mediante la elaboración de Tecnosoles con diferentes aplicaciones ambientales". *Aguas & Residuos* **5**, 12-25.
34. Blakemore, L. C.; Searle, P. L.; Daly, B. K., (1977): *Methods for chemical analysis of soils*. N.Z. Soil Bureau Scientific Report lOA: A 11.1-11.7.
35. Khalid, R.A.,W.H. Patrick, Jr., and R.D. DeLaune. (1977). "Phosphorus sorption characteristics of flooded soils". *Soil Sci. Soc. Am. J.* **41**, 305-310.
36. Olsen, S.R., and Sommers, L.E., (1982). *Phosphorus. In A. L. Page, et al (Eds.), Methods of Soil Analysis*, 2nd ed. Madison, WI: Agronomy No. 9. Am. Soc. Agronomy 2, 403-430.