

## Constructed Wetlands Filled with Dewatered Sludge from a Water Treatment Plant Applied as a Substrate for Phosphorus Removal

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### Highlights:

- The constructed wetlands with Water Treatment Plants (WTP) sludge demonstrated superior phosphorus removal efficiency compared to that with gravel.
- Systems with WTP sludge exhibited significantly lower effluent phosphate concentrations.
- Phosphorus-accumulating organisms that enhance biological removal were identified in systems with WTP sludge.
- Continuous monitoring and substrate renewal are essential for maintaining long-term phosphorus removal efficiency.
- Keywords: Phosphorus removal; Constructed wetlands; WTP sludge.

## INTRODUCTION

Constructed wetlands (CWs) are wastewater treatment systems inspired by natural ecosystems, where nutrient removal occurs naturally. These decentralized and low-cost systems are effective in removing nutrients and organic matter (Von Sperling and Sezerino, 2018). Recently, the potential of dewatered sludge from Water Treatment Plants (WTP sludge) as an alternative substrate has been investigated, especially for phosphorus adsorption, due to its physicochemical characteristics (Zhao et al., 2021).

Phosphorus removal is a crucial aspect of wastewater treatment, as excess phosphorus can lead to eutrophication of water bodies, resulting in algal blooms and deterioration of water quality. Traditionally, phosphorus removal is carried out through chemical and physical processes that can be expensive and generate additional waste. The use of dewatered sludge from WTPs as a substrate in CWs offers a potentially more sustainable and cost-effective solution by repurposing waste from another water treatment process into a valuable resource. Moreover, CWs provide a habitat for a diversity of microbial communities that play essential roles in pollutant degradation and nutrient cycling.

This study aimed to evaluate the efficiency of phosphorus removal in CWs using WTP sludge as substrate. Additionally, the microbial communities present were analyzed using Fluorescence in situ Hybridization (FISH) and 16S rRNA sequencing.

## METHODOLOGY

Six upflow vertical pilot systems were studied over 330 days, named CW1, CW2, CW3, CW4, CW5, and CW6 (Figure 1), the first three (1, 2, and 3) were filled with gravel (in 0, 1, and 3), and the others with dewatered WTP sludge containing Poly Aluminum Chloride (PAC), referred to here as WTP

sludge. The macrophyte planted in systems CW2, CW3, CW5, and CW6 was *Canna indica*; systems CW1 and CW4 were used as controls (not planted) to assess the potential influence of the plants on nutrient removal. Systems CW2 and CW5, as a differential, received micro-aeration in the last 20 cm, thus creating an aerobic zone at the effluent outlet.

These systems operated under different hydraulic retention times (HRT) of 8, 5, 3, and 1.5 days. Weekly monitoring chemical oxygen demand (COD) and ammoniacal nitrogen (N-NH<sub>3</sub>), and monthly monitoring of total phosphorus (TP) and phosphate (PO<sub>4</sub><sup>3-</sup>) concentrations was conducted to evaluate phosphorus removal efficiency. FISH assay was performed as described in Faria et al. (2021).

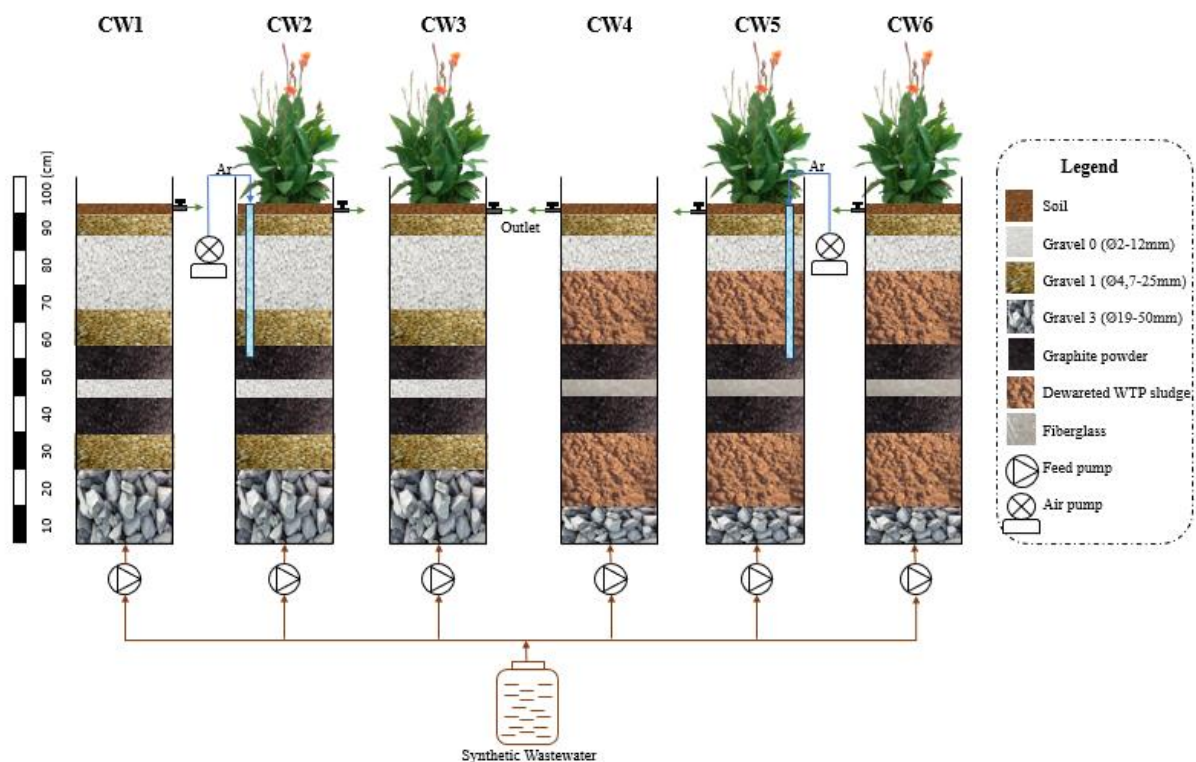


Figure 1. Schematic layout of the different Constructed Wetlands.

## RESULTS AND CONCLUSIONS

The COD removal rates were 91.5% for the gravel and 88.7% for WTP the sludge. For N-NH<sub>3</sub>, the systems with aeration at 0.8 mgO<sub>2</sub> L<sup>-1</sup> min<sup>-1</sup> produced an effluent with low concentrations, below 10 mg L<sup>-1</sup>. The results indicated that systems with WTP sludge had better phosphorus removal rates. The concentrations of PO<sub>4</sub><sup>3-</sup> in the effluent from systems with gravel ranged from 2.73±0.90 mg. L<sup>-1</sup> to 7.40±6.65 mg. L<sup>-1</sup>, whereas in systems with WTP sludge, the results ranged from the limit of quantification (LQ) to 0.79±1.57 mg.L<sup>-1</sup>. Similar results were observed for TP; in the effluent from the gravel modules, the concentrations of TP varied from 5.18 ± 0.86 mg. L<sup>-1</sup> to 7.08 ± 1.87 mg. L<sup>-1</sup>, while in systems with WTP sludge, they ranged from 0.37 ± 0.22 mg. L<sup>-1</sup> to 0.64 ± 0.27 mg.L<sup>-1</sup>.

For HRTs of 5 and 3 days (Table 1), CW4, CW5, and CW6, which were all filled with WTP sludge, showed significantly greater TP removal efficiencies than did CW1, CW2, and CW3, which were filled only with gravel. With a reduction in the HRT to 1.5 days, a decrease in efficiency was observed in the systems, especially those filled with WTP sludge (CW4, CW5, and CW6). This may indicate that the systems reached the adsorption capacity limit of the WTP sludge and/or that system clogging occurred.

Table 1: Total Phosphorus Removal Efficiencies (in %)

HRT	WC1	WC2	WC3	WC4	WC5	WC6
5 days	39,6±36,4	49,8±23,0	39,3±20,5	99,0±3,0*	98,9±3,2*	98,5±4,7*
3 days	16,0±20,8	28,7±19,8	34,8±23,9	87,4±30,8*	100,0±0,0*	100,0±0,0*
1,5 days	21,9±37,9	47,4±22,6	65,0±5,2*	39,5±25,8	71,3±8,7*	57,6±14,4

\*There was a statistically significant difference in the mean comparison test (95% significance level).

FISH analysis (Figure 2) revealed the presence of phosphorus-accumulating organisms (PAOs) in the systems with WTP sludge, which contributed to the biological removal of phosphorus. However, 16S rRNA analysis has not provided much information on phosphorus-accumulating microorganisms. Only two genera were identified: *Devosia* and *Dechloromonas*. Recently, Ni et al. (2022) analyzed new genera related to PAOs and reported that few of these organisms have been identified. Therefore, we suppose that some of the microorganisms found in this study might also have this function. However, they have not yet been reported, or the existing PAOs might belong to another kingdom, such as fungi such as *Saccharomyces cerevisiae*, *Candida humicola*, *Hansenula fabianii*, and *Hansenula anomala* (Akram et al., 2022).

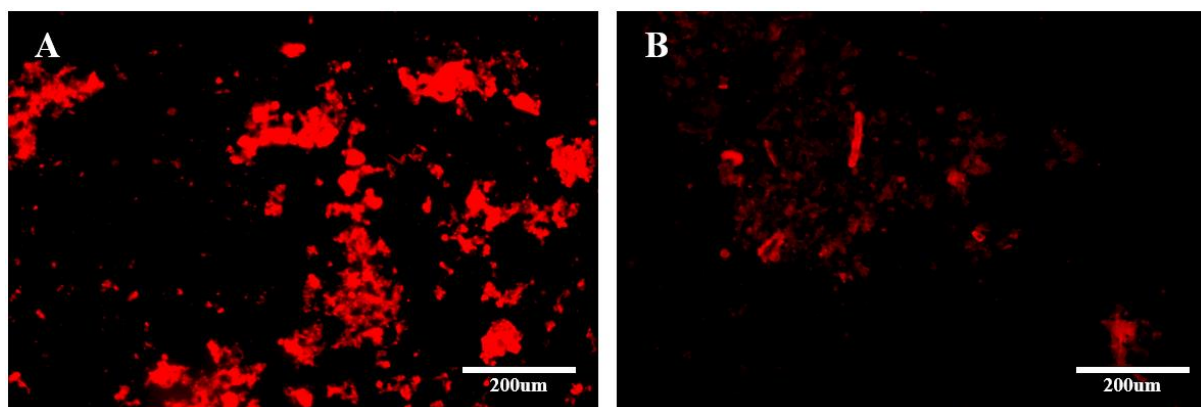


Figure 2. FISH microscopy images of PAOs. A) System with WTP sludge, B) System with gravel.

This study highlights the superior capacity of WTP sludge for phosphorus adsorption compared to that of gravel. The results indicate that systems filled with WTP sludge exhibit significantly greater phosphorus removal efficiency. However, it is essential to continuously monitor the phosphorus levels in the effluent to determine the need for substrate renewal over time, thus ensuring that the system efficiency is maintained. Additionally, future studies should investigate the saturation mechanisms and potential substrate regeneration strategies to extend the lifespan of these systems.

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