Title
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Pruning stormwater biofilter vegetation influences water quality improvement differently in *Carex appressa* and *Ficinia nodosa*.

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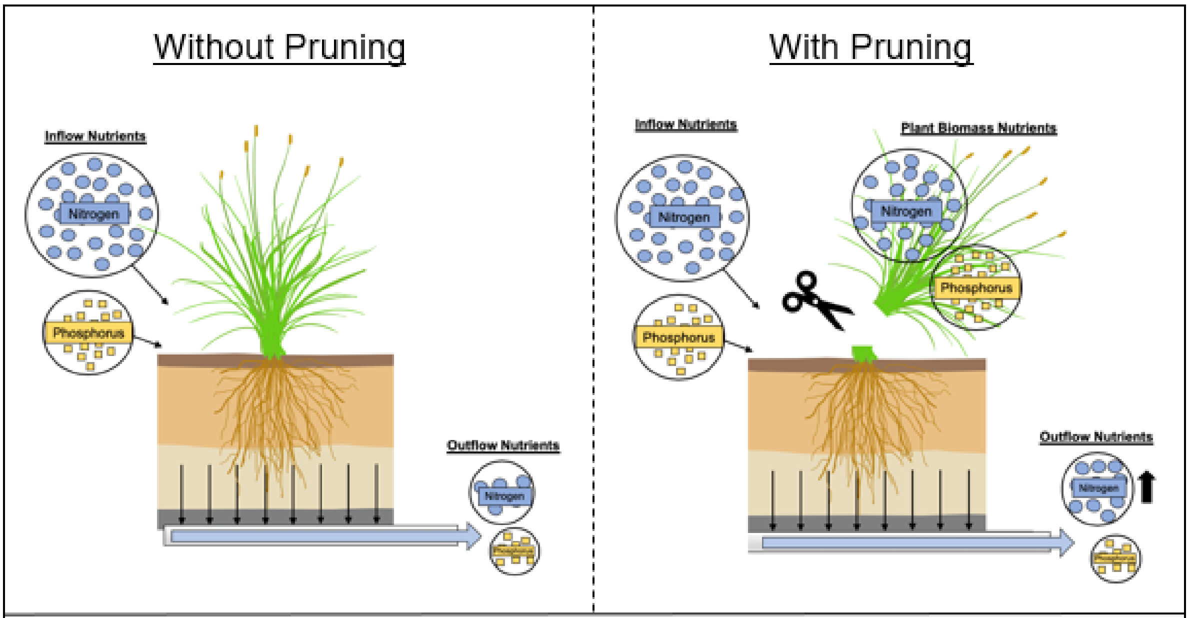
Graphical Abstract



$$Outflow\ Nitrogen_{non-pruned} < Outflow\ Nitrogen_{pruned}$$

$$Nutrients\ Removed\ by\ Columns < Pruned\ Biomass\ Nutrients$$

Abstract

The maintenance of stormwater biofilter vegetation is conducted under local guidelines, which often include seasonal pruning. However, the effects that pruning has on water quality improvement remain unknown. This study used experimental columns to investigate the effects of pruning on effluent concentrations of nitrogen, phosphorus, and metals when planted with two common biofilter plant species, *Carex appressa* and *Ficinia nodosa.* Effluent was monitored in pruned, non-pruned, and unplanted control columns during a 70-day regrowth period, with monthly composite water sampling encompassing the flushed saturated zone water and effluent of each column to best represent a biofilter during a storm event. Differences between pruning treatments and the control were often species-specific and varied with nutrient type. No significant differences between treatments were found for total phosphorus, but pruning treatments affected nitrogen oxide removal in later sampling dates for *F. nodosa*, but not *C. appressa.* Total N and P removal ranged from 77–88% and 66–93%, respectively, by both pruned and non-pruned plants. The overall amount of N and P removed in the pruned biomass was 2.1 – 3.5 times more than the estimated amount removed from the influent by the regrowth of the pruned columns alone during the regrowth period. Consequently, the amount of nutrients removed via pruning may significantly impact long-term removal. Cadmium, copper, lead, and zinc effluent concentrations were similar between treatments with removal efficiencies over 95%. Overall, pruning appears to affect water quality improvement, but optimal pruning practices that may enhance long-term removal should be investigated further.

Keywords: biofilter; best management practice; water sensitive urban design; landscape maintenance; biomass harvesting

1 Introduction

Stormwater runoff becomes increasingly contaminated with both dissolved and particulate contaminants as it traverses the urban landscape (Phillips et al., 2018). These contaminants, without removal, are eventually released into downstream water courses. Excess nutrients are likely to cause algal blooms and eutrophication and additional heavy metals can critically affect living organisms within the system (Phillips et al., 2018, Gunawardena et al., 2015).

Water Sensitive Urban Design (WSUD) systems, also known as Low Impact Development systems and Sustainable Urban Drainage systems (Bratieres et al., 2008), seek to make the way water moves through the urban landscape more closely mimic natural hydrology. One system commonly employed is the stormwater biofilter. Biofilters help remove contaminants from urban stormwater runoff by allowing captured water to percolate through vegetated filter media beds to enhance the sorption of contaminants and to promote biological uptake in both plants and in their rhizosphere (Bratieres et al., 2008).

Biofiltration systems are being increasingly used to combat water contamination in urban areas, but their maintenance requirements, and how to ensure systems are most effective, requires further investigation (Erickson et al., 2018, Delgrosso et al., 2019, de Macedo et al., 2017). Considered designs are also essential to reduce pollutant loading, with not all media- and plant-types being equal. Vegetated biofiltration systems typically remove nutrients more than non-vegetated systems (Henderson et al. 2007; Lucas and Greenway, 2008), leading many researchers to investigate the relative abilities of plant species to further increase removal efficiency (Bratieres et al., 2008, Read et al., 2008, Payne et al., 2014b). Biofilter vegetation may also offer several other benefits, such as wildlife habitat, pollinator services, and biodiversity support (Le et al., in review). The various roles of biofilter plants have been summarised in Dagenais et al. (2018). However, it is also important to ensure that biofilters do not become long-term sources of pollutants. Hatt et al. (2007) noted that non-vegetated soil-based columns were potential sources of pollutants, especially nitrogen, while sand-based columns likely converted particulate nitrogen compounds into dissolved forms. Studies have also shown that vegetated columns can become nutrient sources, with vegetation selection critical to biofilter performance (Bratieres et al., 2008, Payne et al., 2014).

Most of the roles that plants have on biofilter function are likely impacted by the common maintenance practice of pruning (Payne et al., 2015, Erickson et al., 2018). This practice is intended to increase visibility for pedestrians and motorists, and to enhance aesthetics, but it may also offer an additional opportunity to increase pollutant removal efficiency in biofilters (Davis et al, 2006). Plant biomass pruning has been investigated previously for nutrient removal in constructed wetlands (Fogli et al., 2014, Vymazal, 2007, Graber and Junge-Berberovic, 2008) and for specific nutrients and elements (Kim and Geary, 2001, Vymazal et al., 2010), but has still been identified as a key area that is needed in biofilter research (Muerdter et al., 2018, Davis et al., 2006, Roy-Poirier, 2009), and is important for the implications on urban biofilter maintenance and efficiency.

This study seeks to build upon previous research on the capabilities of pruned plants to increase pollutant removal in constructed wetlands and extend it to urban stormwater biofilters using two well-known biofilter plant species. The aim of the current study is thus to determine the impact that pruning vegetation has on water quality improvements in stormwater biofilters, and whether this impact is species dependent. These findings will help landscape and WSUD asset managers determine appropriate maintenance practices for stormwater biofilter vegetation.

2 Methods

Typical vegetation maintenance practices in stormwater biofilters were replicated in a lab-based column study by pruning mature vegetation. To determine the effects of these practices, nutrient and metal concentrations were measured in the effluent of columns with pruned and non-pruned vegetation, as well as non-vegetated control columns. These effects were evaluated on two plant species which are commonly found in southeast Australian stormwater biofilters, *Carex appressa* and *Ficinia nodosa* (Winfrey et al., 2018). The column design, watering regime, and synthetic stormwater runoff recipe used in this experiment were based on previous studies (e.g., Bratieres et al. (2008) Payne et al. (2014b), Read et al. (2008), and Payne (2013)).

2.1 Column Setup

PVC columns (150-mm) were capped on one end and filled with layers of gravel, coarse sand mixed with sugar cane mulch (5% by volume), and filter media (sandy loam, according specifications in Payne et al. (2015)). Media was added to columns 2 L at a time then packed using a slide hammer and plate in order to compact layers evenly. A saturated zone was included to promote denitrification (Figure 1), using organic sugar cane mulch (Brunnings, Australian Certified Premium Product AS4454) as the carbon source. Ten plants of each *C. appressa* and *F. nodosa* were planted individually in columns and allowed to establish for ten weeks in a covered shade house under ambient outdoor conditions with twice weekly watering using tap water (i.e., establishment period). Following the establishment period, the averages of the five tallest stems of *C. appressa* and *F. nodosa* were 707 mm and 583 mm, respectively. At this point, the planted columns, along with five unplanted columns were placed under fluorescent grow lights with a 12-hr photoperiod (daily photosynthetic photon flux density ~ 20 mol/m^2) indoors to mature while being dosed twice weekly with 4.2 L of semi-synthetic stormwater runoff representing typical Melbourne runoff water quality (Table 1) and volume (2% of drainage area) for the duration of the maturation and experimental phases (as described in Payne (2013)). Tap water was used as the source water for the semi-synthetic stormwater runoff, to which we added nutrient and metal salts, dissolved Cd, and sieved (300-µm) sediment from a stormwater pond to adjust tap water pollutant concentrations to the levels shown in Table 1. Target pollutants were previously quantified in the source water by Payne (2013). With the exception of Cu, all pollutants were below the level being targeted in the synthetic stormwater runoff levels. For instance, elevated phosphorus levels are not shown, as phosphorus is not added as a corrosion inhibitor to the mains water in the City of Monash (Yarra Valley Water, 2015). Additionally, we added 0.8 mg/L of sodium thiosulfate pentahydrate ($Na_2S_2O_3.5H_2O$) to dechlorinate the tap water, as described in Payne (2013). Synthetic stormwater runoff was prepared and stored in a 500L mixing tank for each dosing event. All columns were allowed to mature while being dosed with semi-synthetic stormwater runoff for three months prior to applying the pruning treatment in January 2015. Five plants of each species were pruned to 100-mm above the filter media surface and pruned material was removed from the columns. The remaining five plants of each species were not pruned. The five replicates of each of the five treatments were situated in five separate rows (i.e., blocks) underneath rows of grow lights to minimise effects of heterogeneity in environmental conditions in the lab. Each of the five treatments contained five replicates.

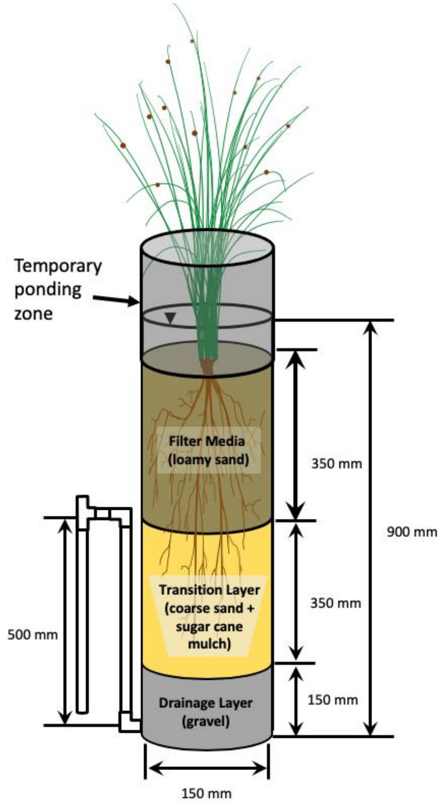


Figure 1 Diagram of column showing layer depths and composition. Saturated zone maintained by upturned pipe.

Table 1. Target and measured influent concentrations for each sampling date.

Parameter	Target Concentration (mg/L)	Influent Concentration on Sampling Days (mg/L)			
		Day 1[*]	**Day 13**	**Day 48**	**Day 70**
Total Nitrogen	2.18	2.0	1.6	2.2	2.1
Ammonia	0.34	0.34	0.16	0.36	0.31
Nitrate/nitrite	0.74	0.97	1.2	0.94	0.98
Total phosphorus	0.35	0.34	0.12	0.35	0.32
Filterable reactive phosphorus	0.12	0.19	0.098	0.18	0.13
Cadmium	0.0045	-[**]	-	-	0.003
Copper	0.05	-	-	-	0.51[***]
Lead	0.14	-	-	-	0.11
Zinc	0.25	-	-	-	0.29

[*] refers to number of days after pruning

[**] concentration not measured

[***] copper levels were elevated in source water

2.2 Water Quality

Water quality in the influent and effluent was measured monthly from February to April 2015 in composite samples of influent and composite samples for each column's effluent in 5L HDPE containers. Composite sampling of the influent occurred by collecting 1L of the prepared synthetic stormwater runoff at the beginning, middle, and end of each dosing event, for a total of 3L. Composite sampling of the effluent from each column was undertaken by collecting all effluent flowing from each column after dosing until the flow ceased, approximately 24 hours after the dose was added. The captured flow was therefore typical of flow from a biofilter during a storm event, with a mixture of flushed saturated zone water and effluent captured. Composite effluent samples were analysed for nutrients and metals concentrations.

2.2.1 Nutrients

Total nitrogen (TN) and total phosphorus (TP) were determined from unfiltered influent and effluent water samples using persulfate digestion methods (Hosomi and Sudo, 1986) at the Water Studies Centre, a National Association of Testing Authorities (NATA)-accredited lab at Monash University. Minimum detection limits for TN and TP were 0.02 and 0.01 mg/L, respectively. Nitrogen oxide anions (nitrate/nitrite; NO_x), ammonia (NH_3), and filterable reactive phosphorus (FRP) were analysed in samples filtered through a 0.45-µm polyethersulfone syringe filter (Sartorius, Germany). Nutrient ion concentrations were determined using flow injection analysis in a NATA-accredited lab according to APHA (1998). Minimum detection limits for NO_x, NH_3, TP, and FRP were all 0.001 mg/L.

2.2.2 Metals

Metals concentrations were measured once before pruning on the last sampling day, and 70 days after pruning. As with nutrient samples, subsamples were collected in 100-mL HDPE bottles, representing influent from the source water and effluent from each column. Samples were acidified before analysis using EPA Method 6020A. Cadmium, copper, lead, and zinc were analysed in water quality samples using an ICP-MS at Australian Laboratory Services (Scoresby, VIC). Minimum detection limits for Cd, Cu, Pb, and Zn were 0.002, 0.01, 0.01, and 0.01 mg/L, respectively.

2.3 Biomass

2.3.1 Plant Growth and Pruned Biomass

The aboveground plant biomass was measured for five individuals randomly selected from the planting stock of each species, with plants sacrificed to estimate initial biomass. Aboveground biomass of all plants was also measured at the end of the experiment and of the pruned material from both species at the time of pruning. Biomass samples were dried at 60°C for 24 hrs then weighed. This was repeated until weight remained unchanged from previous recordings (typically within 48 hours).

To determine whether the 70-day regrowth period was long enough for pruned plants to regrow most of their lost biomass, the final aboveground dry biomass of non-pruned plants was compared to the sum of pruned dry biomass and final aboveground dry biomass of pruned plants.

Stem heights were also measured periodically, averaging the tallest five stems. Stem growth rate was calculated by dividing the difference in stem height averages between two dates by the number of days between sampling.

2.3.2 Tissue Nitrogen and Phosphorus Content

Fresh leaves of both plant species were collected before and after pruning and dried as described previously. Three replicate samples of 2 g dried leaf were ground and homogenised in a ball mill grinder and passed through a 2-mm sieve before analysing on an ANCA GLS2 elemental analyser with a SerCon 20-22 mass spectrometer for nitrogen content. We did not determine phosphorus content of leaves here. Phosphorus content of *C. appressa* leaves were assumed to be 2.7 mg P/g dry biomass, similar to that of the morphologically similar *C. fascicularis*, as in Browning and Greenway (2003). Phosphorus content of *F. nodosa* leaves were assumed to be 3.6 mg P/g dry biomass, similar to that of the morphologically similar *F. trispicata*, as in Basic (2015).

The amount of nitrogen and phosphorus removed in the pruned biomass was calculated by multiplying the N and P content by the dry weight of pruned biomass.

2.4 Data Analysis

Effluent concentrations are primarily used for treatment comparisons, rather than the more typical percent removal, due to influent concentrations varying between sampling events (Table 1). Additionally, effluent concentrations are typically regulated by governments and will ultimately affect downstream watercourses. Effluent concentrations of each pollutant were compared statistically for each sampling event separately. We used the Shapiro-Wilk test to determine normality of raw and log-transformed effluent concentrations for each treatment, pollutant, and sampling event . When normality could be assumed, we tested for equal variances using the Bartlett test. When both normality and equal variances could be assumed, one-way ANOVAs were used to determine significant differences of means. When ANOVAs were significant, post-hoc analyses were completed using Tukey's honestly significant difference tests between treatments. When normality could not be assumed, non-parametric Kruskal-Wallis tests were used to determine significant differences of means. When Kruskal-Wallis tests were significant, post-hoc analyses were completed using pairwise Wilcoxon signed-rank tests between treatments. All tests used a significance value, α, of 0.05.

Daily stem growth rate, measured as stem height (mm), was used as a proxy for the relative growth rate, to help indicate what stage of growth the plant is in. These growth rates were not compared statistically but are tabulated and discussed in relation to effluent concentration differences (see Section 1.3). Final aboveground biomasses were compared within species between pruned and non-pruned treatments using Student's t-tests.

The total amounts of TN and TP removed by the planted and unplanted columns, henceforth referred to only as columns, over the regrowth period was estimated as the product of the total influent volume (84 L) and the differences between average influent and effluent concentrations during the regrowth period for TN and TP, respectively. This approach may have underestimated removal by not accounting for water lost through evapotranspiration (e.g., less TN and TP may have left the columns than our estimate because the effluent volume was likely less than the influent volume). The estimated TN and TP removed by columns was compared to the amount of N and P removed in the pruned biomass during the regrowth period, as determined by the amount of pruned biomass and N and P content.

Statistical tests were not conducted for metals samples since most effluent concentrations fell below detection limits. Percent removal efficiencies were calculated for metals samples when effluent concentrations were above detection limits.

3 Results and Discussion

Most data were not normally distributed ($p > 0.05$, Shapiro-Wilk test), but two ANOVAs could be completed on normally distributed data with equal variances, which occurred for TN one day after pruning and NH_3 70 days after pruning. All other comparisons used non-parametric testing and resulted in an additional 8 groups of comparisons: TN effluent concentrations 48 and 70 days after pruning; NO_x effluent concentrations during all 4 sampling events; and FRP effluent concentrations 13 and 48 days after pruning (Figure 2 and Figure 3).

No significant differences among planted treatments were observed on the first sampling date, one day following the pruning treatment (Table 2; Figures 2 and 3), but significant differences between the unplanted controls and pruned *C.appressa* for total nitrogen and between the unplanted control and NO_x planted treatments did occur. Effluent concentrations of NO_x were significantly lower from planted columns than from unplanted columns through most of the experiment. However, during the last two sampling events, effluent NO_x concentrations from columns planted with pruned *F. nodosa* were no different to unplanted columns (Table 2; Figure 2), potentially due to plant stress or altered physiological conditions. Metal removal was generally very high in both pruned and non-pruned treatments and most effluent concentrations were below detection limits.

In both species, there were no significant differences between the final aboveground biomass of non-pruned plants and the sum of pruned biomass and final aboveground biomass for pruned plants ($p > 0.05$), indicating that full regrowth was achieved in 70 d.

3.1 Nitrogen

Total nitrogen (TN), ammonia (NH_3) and nitrogen oxide anions ($NO_3^- + NO_2^-$) were analysed for pruned and non-pruned samples of *C. appressa* and *F. nodosa* and the unplanted control columns. TN percent removal ranged from 77-88% throughout the study for the vegetated columns, indicating a net sink of nitrogen. This relatively high N removal efficiency has been demonstrated in previous studies, as summarised by Payne et al. (2014a). Significant differences in TN removal were observed on multiple sampling dates. Columns planted with *C.appressa* generally provided better TN removal than those planted with *F.nodosa* (Table 2 and Figure 2a). Pruned *C.appressa* were able to significantly reduce effluent concentrations as compared to the unplanted control for sampling days 1, 48, and 70 days after pruning, while non-pruned *C.appressa* only had significantly lower concentrations than the unplanted control for the last two sampling dates (Days 48 and 70). *F.nodosa* only had significantly better TN removal compared to the unplanted control for the last sampling day for the non-pruned treatment, with the pruned treatment consistently performing similar to the unplanted control.

As is typical of new plant growth, the N content of tissue of recently pruned plants in this experiment was higher than non-pruned plant N content (Greenwood, 1976). The difference between plant tissue N content of pruned and non-pruned samples was greater in *C. appressa* than in *F. nodosa*. After 48 days, the plant tissue N content, represented as % (w/w), for *C. appressa* was 1.61% and 0.87% for pruned and non-pruned samples, respectively, while for *F. nodosa* it was 0.90% and 0.75%. After 70 days, this difference was not as large, especially for *F. nodosa*, with pruned and non-pruned plants sharing almost the same tissue N content (0.73% and 0.71%, respectively). The percent difference for *C. appressa* was still noticeable after 70 days however, with pruned and non-pruned samples holding 0.95% and 0.76% plant tissue N content. This sudden increase in plant tissue N in *C. appressa* after pruning is likely due to the increase in fresh biomass that was grown, as indicated by the daily stem growth rates (Table 3). The increased tissue N content of fresh biomass in new leaves growing from the pruned *C. appressa* may have allowed pruned plants to remove N at a similar rate to the non-pruned columns, which had a larger stock of standing biomass and thus higher water uptake rates. Therefore, it could be expected that the pruned plants were able to take up more N on a per weight basis than non-pruned plants. This counter levering of nitrogen between higher N content fresh biomass in the pruned treatment and a larger quantity of non-pruned biomass in the non-pruned plants may explain the similarity of TN effluent concentrations for the two *C. appressa* treatments.

This is also supported by nitrogen removal in the *F. nodosa* samples, as the smaller differences in nitrogen content between pruned and non-pruned plants, as well as the more comparable stem growth rates (Table 3), did not result in the pruned samples improving water quality over non-pruned samples and, in fact, resulted in higher TN effluent concentrations on the final two sampling dates (Table 2; Figure 2a). This thus helps to show that not only is choosing the most efficient plant species for water quality improvement important (Kim and Geary, 2001, Read et al., 2008, Bratieres et al., 2008), but how they are maintained may also affect water quality. Given the much smaller difference in growth rate between pruned and non-pruned samples of *F. nodosa* than of *C. appressa*, the more significant results for *F. nodosa* may be due to an inappropriate pruning method or time. Muerdter et al. (2020), in a study focussing on the nutrient removal capabilities of three plants, found that removing shoots through harvesting would drastically decrease the biomass nitrogen content of the system, but did not determine the impacts that this level of harvesting would have on the regrowth capabilities of the plants. Therefore, the management of polycultures in biofilters should take each species into consideration before maintenance, such as pruning, is undertaken.

*Table 2 Significance values using ANOVA and Kruskal-Wallis results comparing unplanted control, pruned vegetated columns, and non-pruned vegetated columns for each sampling day for the five nutrient categories. Bold indicates $p < 0.05$. *Indicates ANOVA results were used; all other values from the Kruskal-Wallis test.*

Significance Values				
Days since harvest	**1**	**13**	**48**	**70**
Total nitrogen	**0.026***	0.087	**0.002**	**0.001**
Ammonia (NH_3)	0.082	0.337	0.170	**0.003***
Nitrogen oxide anions (NO_x)	**0.005**	**0.012**	**0.001**	**0.000**
Total Phosphorus	0.143	0.110	0.473	0.328
Filterable reactice phosphorus	0.216	**0.002**	**0.001**	0.199

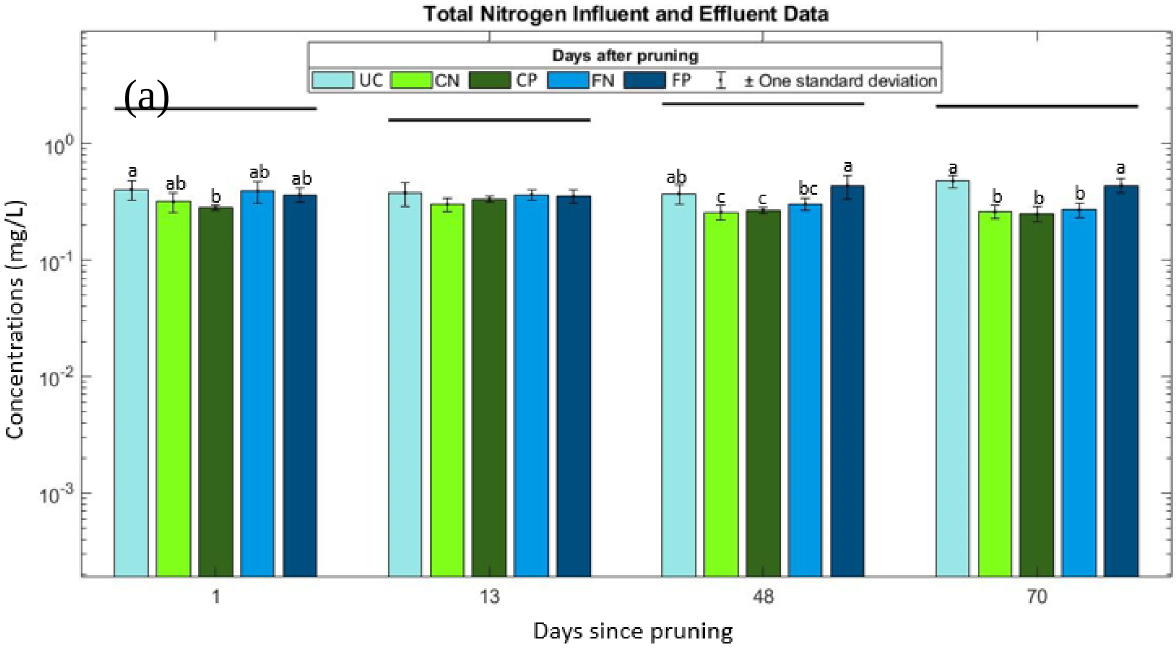
Ammonia removal was generally high in all columns, with overall removal rates ranging from 87-96% for planted treatments. Neither *C. appressa* nor *F. nodosa* showed significant differences between pruned and non-pruned samples, nor the unplanted controls. Only columns with non-pruned plants removed ammonia better than unplanted controls and only on the sampling date 70 days after pruning (Table 2 and Figure 2b). Bratieres et al. (2008) noted that even non-vegetated biofilters were able to remove >93% of ammonia from stormwater. However, biofilters with healthy plants may be able to maintain removal during dry periods when nitrification is diminished while non-vegetated biofilters may not support nitrifying microbial communities (Glaister et al., 2014).

For nitrogen oxides, all planted treatments performed significantly better than the unplanted controls, except for the pruned *F.nodosa* 48 and 70 days after pruning, which had the same removal as unplanted controls (Figure 2c). Hydraulic short-circuiting of biofilters may occur as a result of root dieback increasing hydraulic conductivity in the filter media (Archer et al., 2002); this could result in nitrate reaching the column outflow during sampling events. In the final two water quality sampling events, NO_x effluent concentrations were much higher in columns with pruned *F. nodosa* (Figure 2c), with an average percentage removal rate between 85.0%-86.7% as opposed to a minimum of 99.5% for all other dates, and the effluent concentration exceeding the unplanted control 48 days after pruning. These sampling dates correspond to a marked decline in stem growth rate of pruned *F. nodosa*, which occurred between 41 and 50 days following pruning (Table 3). Similarly, Greenway and Lucas (2010) pruned vegetation in experimental biofilter mesocosms containing, among other species, *C. appressa* and *F. nodosa*, noting that *F. nodosa* did not react favourably to pruning. This decline in growth rate may have indicated plant stress and, consequently, resulted in root dieback (e.g., Todd et al. (1992)) that allowed higher amounts of NO_x to reach the outflow during sampling events. However, different plant species respond differently to pruning (Thorne and Frank, 2009), some of which may not experience any root dieback following pruning (Balogianni et al., 2014). In this case, the differences in NO_x removal between pruned and non-pruned *F. nodosa* may have been related to altered plant physiological processes rather than hydraulic short-circuiting. Saifuddin et al. (2010) investigated different pruning strategies on *Bougainvvillea glabra* and noted the negative impact complete pruning had on shoot and root growth, but that complete, partial, or frequent pruning often gave better results for other measures (quantum yield, chlorophyll values, etc.). If pruning *F. nodosa* did diminish plant growth and transpiration was reduced, then NOx removal could have decreased due to less plant uptake, which is often the most important N removal mechanism when plants with high N uptake are present (Morse et al., 2018), such as *F. nodosa* (Read et al., 2008). Pruning vegetation can affect soil nutrient cycles by increasing soil respiration and altering microbial community activity (Antonsen and Olsson, 2005). Although this study did not likely last long enough to capture the potential effects of decreased litterfall due to pruning, the removal of this soil carbon input could impact nutrient processing over the long-term by affecting soil faunal communities and microbial nutrient cycles in stormwater biofilters (Mehring and Levin, 2015, Mehring et al., 2016).

The overall amount of N removed by columns during the regrowth period, which was based on the product of the total influent volume (84 L) and the difference between average influent and effluent concentrations, was 155 and 140 mg N for pruned *C. appressa* and *F. nodosa* columns, respectively. The total amount of N removed in the pruned biomass, which was based on the dry weight of biomass and tissue N content, was 388 and 298 mg N for *C. appressa* and *F. nodosa*, respectively, demonstrating that the N removed in pruned material represents a significant component of N removal. If N removal was maintained at the same efficiency over the course of the year, annual pruning may account for roughly 30% of annual N removal.

Table 3 Daily stem growth rates for pruned and non-pruned plants post-pruning in mm/day

	Stem growth rates (mm/day)					
Days since pruning	**6**	**13**	**27**	**41**	**50**	**70**
***C. appressa* pruned**	11.7	18.8	15.9	15.9	12	9.4
***C. appressa* non-pruned**	4.6	3.4	0.4	0.8	1.2	0.3
***F. nodosa* pruned**	3.9	6.1	5.2	5.3	4.0	4.6
***F. nodosa* non-pruned**	4.5	4.3	3.6	3.8	3.6	3.1


Total Nitrogen Influent and Effluent Data
Days after pruning
UC
CN
CP
FN
FP
± One standard deviation
(a)
10^0
10^-1
10^-2
10^-3
Concentrations (mg/L)
a
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Days since pruning

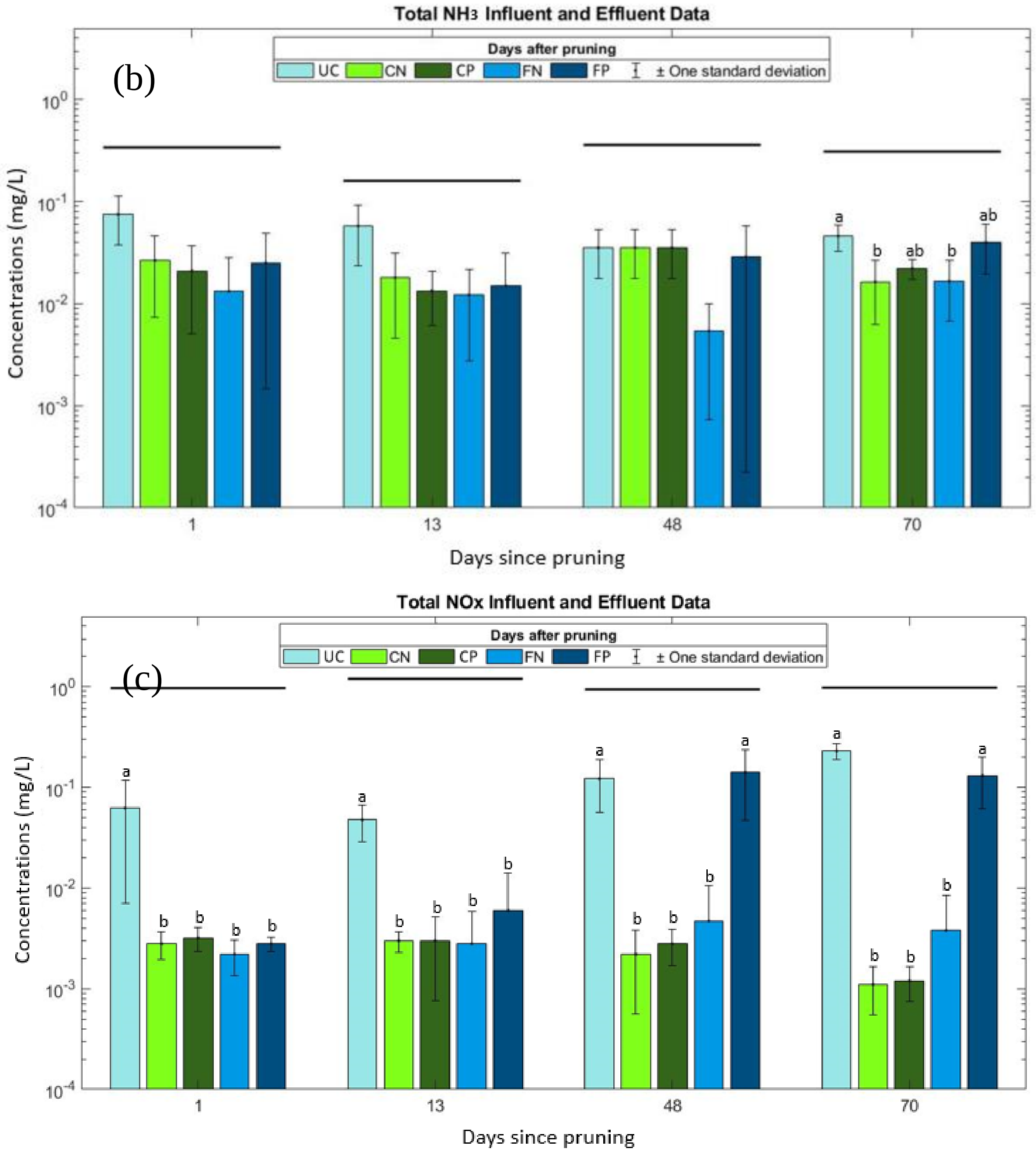

Figure 2 Bar charts showing effluent concentration observations of (a) total nitrogen (TN), (b) ammonia (NH3), and (c) nitrogen oxide anions (NO_X) for the four sampling dates. The influent of each treatment is shown as a horizontal black line. A log-transformed plot was used to maintain the same axes for each nutrient. Five replicates for each mesocosm (N=5). Different letters within the sampling event days indicate a significant difference between treatments.

3.2 Phosphorus

Total phosphorus (TP) and filterable reactive phosphorus (FRP) were analysed for pruned and non-pruned samples of both plant species, as well as an unplanted control. Figure 3 details the effluent concentrations for the sampling dates after the biomass was harvested.

No significant results between planted and unplanted mesocosms were found for TP (Table 2). TP percent removals were also considerably lower in the second sampling date (68.3%-71.7%) than all other sampling times (88-92%); however, this was demonstrated across all samples, and was likely a mathematical artefact of the lower influent concentration observed on the second sampling date. The system is thus a net sink of phosphorus

Phosphorus is one nutrient that has been specifically targeted in previous studies as a nutrient which would benefit from pruning to remove it from the immediate area (Davis et al., 2006). Kim and Geary (2001) state that only 5% of phosphorus is removed by plant harvesting in their constructed wetland study, but indicate that Reed et al. (1995) found a direct correlation between harvesting and phosphorus removal; within certain constraints, phosphorus uptake by the macrophytes in their study increased up to 20-30% of initial influent values. This is an important difference, as the increased uptake of dissolved phosphorus through biological and microbial processes (Muerdter et al., 2018) decreases the total amount of phosphorus entering downstream waterways. Although phosphorus removal is largely dependent on the filter media (Davis et al., 2009) rather than plant uptake, as can be seen when comparing the unplanted columns with all planted columns in Figures 3a and 3b, plants do have the potential to improve the removal of more bioavailable forms of phosphorus (FRP) (Glaister et al., 2014, Bratieres et al., 2008, Hatt et al., 2009). Although this was not observed in this study, removal of plant biomass offers another permanent removal option to reduce the amount of phosphorus cycling through the system.

Removal efficiencies for FRP remained high throughout the experiment, with the lowest percent removal at 95.5%. On the third sampling date, *F. nodosa*, both pruned and non-pruned samples, performed significantly worse than the unplanted treatment, while *C. appressa* did not perform significantly different to the unplanted treatment(Table 2, Figure 3). Although significant results are shown in Table 2 for the second sampling date, no clear trend in significance was found between the planted and unplanted treatments (Figure 3b). The significant increase in FRP and TP removal within two months of the initial pruning for *F. nodosa* (Figure 3) coincides with the higher growth rates of the plants (Table 3), and corroborates findings by Kim and Geary (2001). However, this also demonstrates the differences between species in nutrient removal capabilities (Kim and Geary, 2001, Read et al., 2008). Several studies note the high nutrient removal ability of *C. appressa* in biofilter column studies, especially for phosphorus compounds (e.g., (Bratieres et al., 2008, Read et al., 2008)). In keeping with these findings, columns planted with *C. appressa* removed almost all FRP entering through the influent and, even though there are no significant differences between most planted treatments with the unplanted treatment (Figure 3b), in practical terms there was little difference between non-pruned, pruned, and unplanted sample removal efficiencies (95.5-97.6% removal).

The overall amount of P removed by columns during the regrowth period was estimated as 20 mg P for both pruned *C. appressa* and *F. nodosa* columns. The total amount of P estimated to be present in the pruned biomass was 52 and 77 mg P for *C. appressa* and *F. nodosa*, respectively, demonstrating that the P removed in pruned biomass represents a significant component of P removal. If P removal was maintained at the same efficiency over the course of year, annual pruning could account for roughly 34 and 43% of annual P removal for biofilters planted with *C. appressa* and *F. nodosa*, respectively. Although phosphorus removal mechanisms are largely associated with filter media, as suggested by the lack of significant differences between planted and unplanted treatments in this study, these results suggest that removing plant biomass does not diminish P removal by the biofilter columns. Consequently, pruning biofilter plants may prolong the time before filter media becomes saturated with P, thus improving long-term efficiency of P removal, as suggested by Davis et al. (2006). Fowdar et al. (2017) suggested that removal of aboveground biomass may enhance P removal during the establishment phase of plant in bioretention, but we did not observe that here.

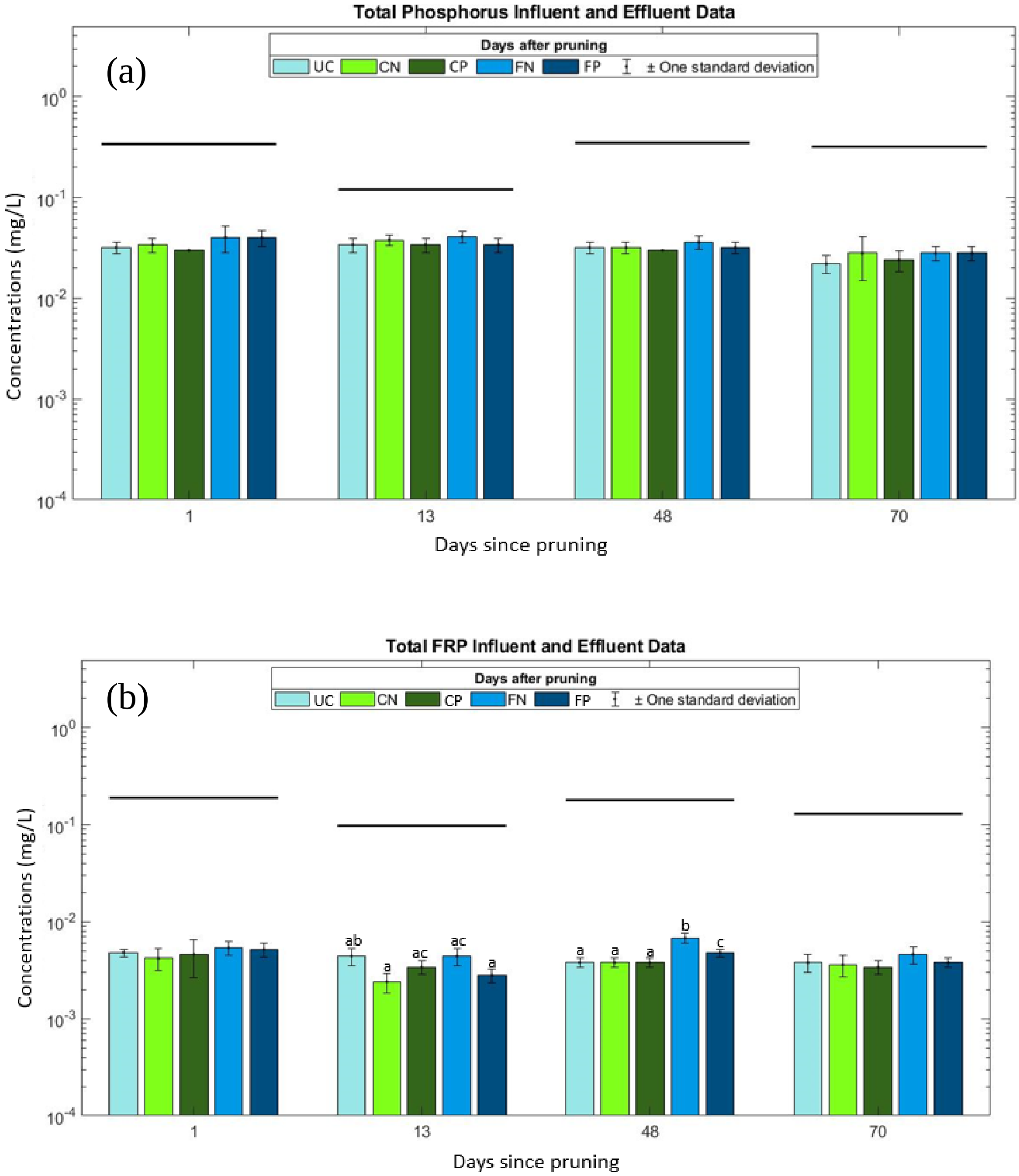

Figure 3 Bar charts showing effluent concentration observations of (a) total phosphorus (TP) and (b) filterable reactive phosphorus (FRP) for the four sampling dates. The influent of each treatment is shown as a horizontal black line. A log-transformed plot was used to maintain the same axes for each nutrient. Five replicates for each mesocosm (N=5). Different letters within the sampling event days indicate a significant difference between treatments.

3.3 Metals

Cadmium, copper, lead, and zinc were analysed both before and after pruning. All effluent concentrations of cadmium, copper, and lead were below detection limits, showing that both pruned, non-pruned, and unplanted mesocosms were able to remove significant amounts of these metals, especially for copper and lead. Zinc removal was slightly more variable throughout the sampling period, however the average removal capacity remained above a 95% removal rate for all treatments. Heavy metal percent removal was similar to previous studies (Muthanna et al., 2007, Blecken et al., 2009).

Plants have been shown to affect metal removal in biofilters (Muthanna et al., 2007, Blecken et al., 2011, Feng et al., 2012), however, pruning did not impact heavy metal removal in this study and metal removal was also unaffected by the presence of plants. Filtering in the surface layers likely removed sediment-bound heavy metals, which has been previously observed (Blecken et al., 2009). Indeed, filter media tends to be the most significant factor in heavy metal removal by biofilters (Read et al., 2008, Sun and Davis, 2007). Nevertheless, it is promising that heavy metal removal did not diminish as a result of pruning if this is to be practiced in biofilters.

4 Limitations/Further studies

In their field studies on runoff regimes and bioretention systems Hatt et al. (2009) noted the challenge of removing nitrogen, given its numerous removal pathways (Vymazal, 2007, Lucas and Greenway, 2008). Biological mechanisms available for nitrogen removal, in comparison to phosphorus removal, involve more microbial activity, and rely more heavily on plant uptake (Muerdter et al., 2018). Maintaining healthy plants may be key for effective nitrogen removal, not just via plant uptake, but also for providing the appropriate conditions for microbial activity in the nitrogen cycle (Dagenais et al., 2018). Further data on the plants used in this study to allow for in-depth plant physiological reactions to pruning to be determined is incomplete; this limits the discussion on how plant uptake mechanisms, including the root systems, are affected by pruning. Further studies into how root systems, plant longevity, and multiple pruning cycles ultimately affect the plants are required for management practices to be specified in-depth. Field studies on pruning biofilter vegetation could determine how frequently and to what extent pruning actually takes place in practice. Mowing grasslands to simulate effects of grazers often increases biomass and species diversity (Turner et al., 1993, Collins et al., 1998, Kitchen et al., 2009). Although highly managed lawns may not increase primary productivity over infrequently managed lawns (Falk, 1980), they may influence partitioning between above- and belowground biomass (Lilly et al., 2015). Additionally, pruning may impact biofilter performance by altering composition of leaf litter, soil carbon sequestration, and soil moisture. These impacts could affect soil faunal communities and nutrient cycling, as suggested by Mehring and Levin (2015). Pruning may be considered an essential practice if the same benefits observed in grasslands are conferred in biofilter plant communities. On the other hand, if biofilter plants receive no benefit from pruning and pollutant removal is marginally impacted, pruning may be unnecessary.

Further studies should measure nutrient effluent concentrations and effluent volume from columns containing plants pruned at different times of the year and growing under appropriate seasonal conditions (e.g., light intensity and duration, temperature, and rainfall patterns). These systems should carry out observations beyond the regrowth period to track differences between pruned and unpruned plants as plants mature. Additionally, belowground plant traits should be measured to determine whether changes to root morphology and belowground nutrient dynamics occur, as observed in grasslands (Kitchen et al., 2009).

5 Conclusions

The impact of pruning on two common Australian biofilter plant species, *C. appressa* and *F. nodosa*, was tested in a lab-based study using semi-synthetic stormwater runoff. These two sedge species are often two of the best-performing plants in terms of nutrient removal. In this study, plant species were just as important as pruning for nutrient removal. For the significant results found, *C. appressa* treatments and non-pruned *F. nodosa* were in general more effective than the non-pruned *F.nodosa* and unplanted treatments, but often there was no pattern to the significant/non-significant treatments. No system within the 70-day regrowth period became a net source of nutrients. Heavy metals had very high removal rates for all columns and were unaffected by pruning.

Nitrogen removal was often better in the planted mesocosms, with non-pruned *F. nodosa* being the exception to this overall trend. The extensive removal processes of nitrogen available in planted mesocosms provide a number of pathways of removal, which may cause some species, under an inappropriate harvesting schedule, to be detrimentally impacted, as occurred with the *F. nodosa*. Phosphorus removal primarily occurs by the filter media through sorption processes and physical straining, of which the former may be maintained when plant uptake increases, or plant biomass is removed.

The results of this study show that pruning plants, for the most part, does not diminish nutrient removal during the regrowth period. Considering the amount of nutrients removed in the pruned biomass, pruning may enhance nutrient removal considerably on a long-term basis. Indeed, when pruned annually and assuming consistent performance year-round, removing plant biomass could account for more than a third of overall N and P removal in stormwater biofilters, respectively. This study also demonstrates the different impacts pruning can have on different plant species, with some impacts rendering the pruned mesocosms no more effective than an unplanted treatment.

Further studies will be required to determine the correct strategy for pruning different species commonly used in biofilters, including pruning length, frequency, and timing.

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Conflicts of Interest

The authors have no conflicts of interest to declare.

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