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System response, nutria herbivory, and vegetation recovery of a wetland receiving secondarily-treated effluent in coastal Louisiana

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ABSTRACT

The City of Hammond, Louisiana began discharging secondarily-treated municipal effluent into Four Mile Marsh in the northwestern Joyce wetlands during fall of 2006. At the time discharge began, these wetlands had been isolated from virtually all freshwater inflow from the surrounding watershed for over a half century, due primarily to the construction of canals and spoil banks. Immediately following effluent discharge in 2006, there was robust growth of herbaceous vegetation. By late fall 2007, the emergent wetlands in the immediate vicinity of the effluent discharge began to decline, and within months nearly the entire marsh south of the discharge pipe had converted to open water or mudflat. By 2010, there had been substantial recovery of the marsh. A number of hypotheses have been presented to explain the degradation of the marsh to open water and mudflats, including herbivory by the introduced rodent nutria (*Myocastor coypus*), excessive nutrients, reductions in above- and belowground biomass, increased soil decomposition due to high nutrients, prolonged inundation, toxicity, increased pH, and disease. Intensive field and mesocosm studies provide conclusive data that the marsh loss was primarily caused by nutria herbivory, and secondarily by waterfowl herbivory, and that recovery of the herbaceous vegetation occurred as a result of aggressive nutria control (>2000 eliminated). Marsh recovery has been most intensive near the point of discharge. Mature baldcypress growing in the area of discharge had growth rates that were five times those of trees not receiving effluent in the lower Joyce area and Maurepas swamp. Field and mesocosm studies show that nutrients increased both above- and belowground biomass and did not increase decomposition rates of herbaceous vegetation. Increased flooding due to lack of drainage from the area is hindering marsh recovery.

1. Introduction

Approximately 30% of the Mississippi River delta plain has been lost since 1956 (Barras et al., 2008; Couvillion et al., 2011), mostly from the lack of seasonal inputs of fresh water, nutrients, and sediments from the Mississippi River (Day et al., 2000). Flooding from the river formed the delta over the past 6000–7000 years but flood control levees built during the last two centuries separated the river from its floodplains, causing prolonged flooding, saltwater intrusion, subsidence, and conversion to open water (Kesel, 1988, 1989; Mossa, 1996; Roberts, 1997; Day et al., 2007). Other factors exacerbating wetland loss include altered hydrology due to the proliferation of dredged canals and deep-well fluid withdrawal associated with the oil and gas industry (Turner et al., 1994; Day et al., 2000; Morton et al., 2002; Chan and Zoback, 2007), intentional impoundment for waterfowl management (Day and Holz, 1990; Boumans and Day, 1994), and herbivory by nutria (Shaffer et al., 1992; Evers et al., 1998).

Baldcypress-water tupelo (*Taxodium distichum-Nyssa aquatica*) swamps once comprised 90% of the wetland habitat in the upper Lake Pontchartrain Basin of southeast Louisiana (Saucier, 1963), which includes the Joyce wetlands. Currently, the vast majority of

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these wetlands are degraded or degrading (Chambers et al., 2005; Shaffer et al., 2009a). To restore this area, a reliable source of fresh water is needed to prevent saltwater intrusion (Thomson et al., 2002) and to increase vertical accretion, through either direct sediment deposition or organic soil formation (Morris et al., 2013a, b; Nyman, 2014). One potential source of fresh water for restoration efforts is disinfected secondarily-treated municipal effluent. Both natural and constructed wetlands have been shown to effectively reduce biochemical oxygen demand, total suspended solids, and nitrogen and phosphorus concentrations (Kadlec and Wallace, 2009; Kadlec and Knight, 1996), and are referred to as 'assimilation wetlands' (Day et al., 2004; Hunter et al., 2009a,b). The benefits of discharging municipal effluent into wetlands rather than the business-as-usual practice of discharging into surrounding rivers and streams include improved water quality (Day et al., 2004), financial and energy savings (Ko et al., 2004), increased primary production (Hesse et al., 1998; Day et al., 2004; Brantley et al., 2008; Hunter et al., 2009a; Lundberg et al., 2011), and enhanced vertical accretion (Rybczyk et al., 2002; Brantley et al., 2008; Hunter et al., 2009b). Increases in aboveground biomass and root production enhances organic soil deposition and carbon sequestration that results in a healthier wetland (Day et al., 2004; Morris et al., 2013a). Geological subsidence of this organic soil results in significant permanent carbon burial. The introduction of treated municipal effluent into highly degraded wetlands of Louisiana is a major step towards their ecological restoration. The following wetland assimilation sites (and quantity of effluent being discharged) are currently functioning in Louisiana: Breaux Bridge (3800 m^3 day^{-1}; Hunter et al., 2009b), Amelia (3000 m^3 day^{-1}; Day et al., 2006), Mandeville (7200 m^3 day^{-1}; Brantley et al., 2008), Thibodaux (11,500 m^3 day^{-1}; Hunter et al., 2009b), Luling (6000 m^3 day^{-1}; Hunter et al., 2009b), Broussard (5700 m^3 day^{-1}) and Hammond (14,500 m^3 day^{-1}; Hunter et al., 2009b), as well as several others. It should be noted, however, that with the exception of the Hammond site (the focus of this analysis), the discharge area at most of the assimilation sites is forested.

In the fall of 2006, the city of Hammond, Louisiana began discharging 11,000–15,000 m^3 day^{-1} of disinfected secondarily-treated municipal effluent into Four Mile Marsh (herein referred to as the Hammond Assimilation Wetland) on the northern border of the Joyce wetlands (Fig. 1). The primary goals of the project were to improve local water quality by wetland filtration and, in the process, to revitalize the hydrologically-isolated and saltwater-influenced wetlands with fresh water and nutrients. However, by late fall 2007 the emergent wetlands in the immediate vicinity of the effluent discharge began to deteriorate, and within months nearly the entire marsh south of the discharge had converted to open water or mudflat. Over half of the marsh had recovered by 2010 and the extent of wetland coverage varied from year to year depending on the extent of *Hydrocotyle* and nutria activity. The area that recovered first and that has most consistently remained as emergent vegetation is nearest to the discharge (Fig. 2).

As can be expected, numerous individuals and environmental organizations began to express concern about the conversion to open water, generating many hypotheses including excessive nutrients, excessive herbivory, reductions in above- and below-ground biomass, increased decomposition due to high nutrients, and disease. Here we test several of these hypotheses and present

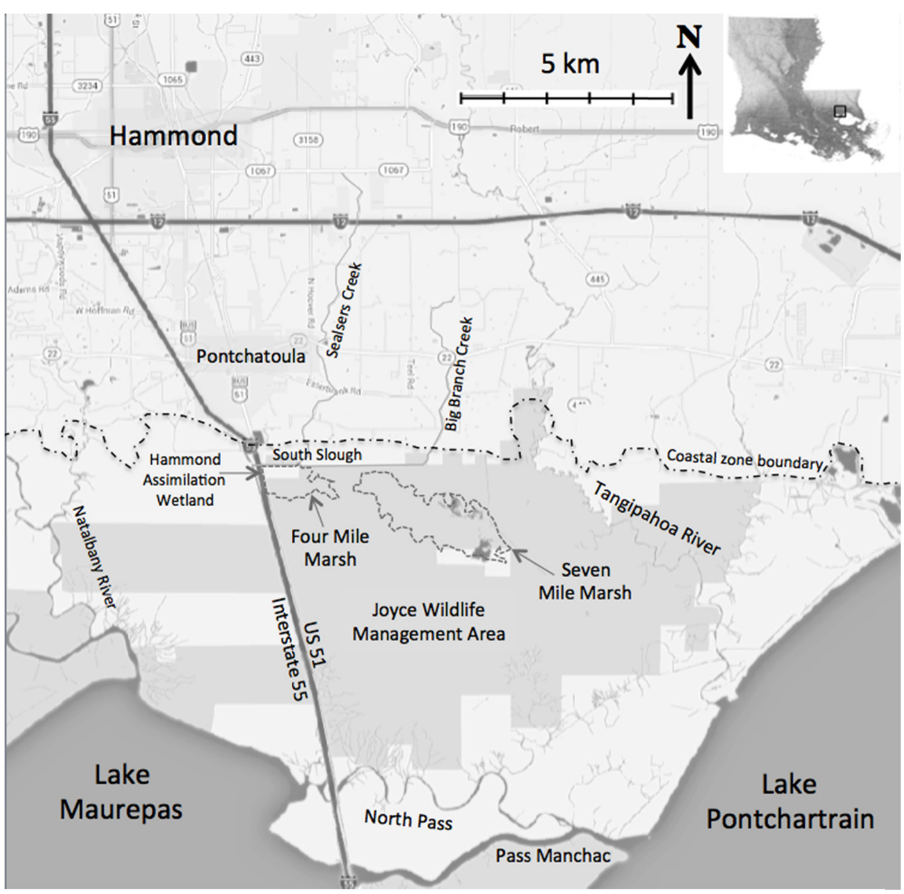

Fig. 1. The Joyce Wetlands, located south of Ponchatoula, Louisiana.

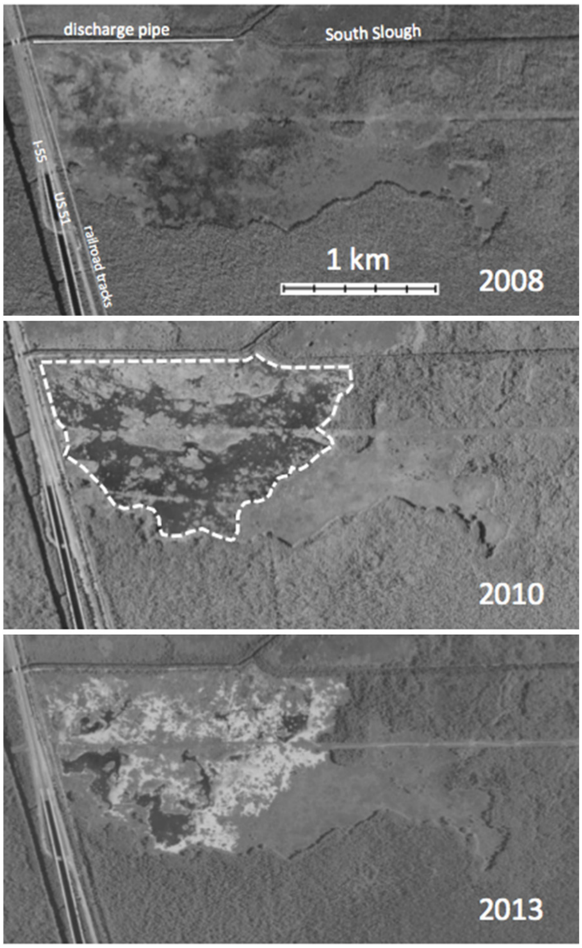

Fig. 2. Satellite images of the Hammond Assimilation Wetland over time. The dashed line indicates the maximum extent of open water in 2008–2009.

suggestions for future activities to prevent such an event from occurring at other wetlands, primarily marsh sites, receiving treated municipal effluent.

2. Objective and hypotheses

The main objectives of this study were to investigate and test the most plausible hypotheses for vegetation degradation at the Hammond Assimilation Wetland.

2.1. Hypotheses tested

1. Excessive nutrients. Inorganic nitrogen and phosphorus concentrations were higher than the assimilation capacity of the wetland and, thus, nutrient overloading caused a decline in vegetation.
2. Baldcypress biomass production. The discharge of treated effluent resulted in low woody productivity, especially near the discharge system.
3. Herbaceous biomass production. The discharge of treated effluent resulted in low herbaceous aboveground biomass production, especially near the discharge system.
4. Nutria herbivory. Herbivory, primarily by nutria, was responsible for most of the marsh deterioration at the Hammond Assimilation Wetland.
5. Waterfowl herbivory. Reduction in herbaceous biomass was also due to grazing by waterfowl.
6. Reduced belowground biomass. The discharge of treated effluent resulted in low belowground biomass, especially near the discharge system, causing vegetation collapse.
7. Increased rates of decomposition. The discharge of treated effluent increased decomposition rates, especially near the discharge system.

3. Materials and methods

3.1. Study site

The Hammond Assimilation Wetland is a part of the Joyce wetlands that extend from North Pass, located north of Pass Manchac, to the Pleistocene uplands that begin south of the city of Ponchatoula (Fig. 1). They are bordered to the east by the Tangipahoa River and to the west by the Natalbany River, but are bisected from north to south by Interstate-55, US-51 as well as railroad tracks. The original hydrology of the Joyce wetlands was characterized by diffuse runoff from uplands directly to the north, and by flow from Selsers and Big Branch creeks, which drain an 86 km^2 watershed that extends north of Hammond. This freshwater input was generally a one-way southerly flow into the wetlands. Water flux into the area from the south was generally tidally driven. A series of well-developed tidal channels along North Pass attest to this tidal influence.

With the exception of Four Mile Marsh and Seven Mile Marsh (Fig. 1), the entire Joyce tract was baldcypress-water tupelo (*Taxodium distichum-Nyssa aquatica*) forest well into the 20th century. Between 1865 and the early 1950s, the baldcypress forest was harvested, including one tree estimated by timber interests to be 4000 years old. The baldcypress trees were first thinned to build a rail line from New Orleans to points north of Lake Pontchartrain (Mancil, 1972, 1980). In 1852, the New Orleans Jackson and Great Northern Railroad built a track across the 'Manchac land bridge,' the isthmus between Lakes Pontchartrain and Maurepas. When the Civil War broke out, the rail line became a strategic target, with the Manchac land bridge, for a time, being the dividing line between the Confederate and Union armies. Much of the railroad was built on wooden bridges, which were torched by both sides. Baldcypress harvesting began shortly after the Civil War. The rebuilt railroad made it easier to move cypress to markets in the Midwest and Northeast. In the 1870s, timber barons began to harvest the gigantic trees, and by the 1890s, steam equipment and pull-boat logging gave the industry more capability to harvest in such difficult terrain (Burns, 1980; Keddy et al., 2007). From the late 1800s through the early 1900s, small logging and farming villages thrived on the land bridge (Keddy et al., 2007). The Louisiana Cypress Lumber Co., milled its last baldcypress log in 1956.

There have been a number of substantial changes over the past century that significantly altered the hydrology of the Joyce wetlands. The earliest impact was the construction of the railroad in the mid 19th century. The railroad was initially built on a bridge spanning the wetlands, but it was rebuilt on the raised embankment that stands today. Although the embankment has a number of small openings for drainage, it severely restricted east-west water movement. US-51 constructed in 1926 on an embankment, parallel to and just west of the railroad, further reduced east-west flow. This was followed by the construction of I-55 in the 1960's, an elevated roadway with a large (60 m) and deep (3 m or more) canal running parallel. This canal now serves as the major drainage channel for the region, shunting water directly into Lake Maurepas, and for the most part, bypassing the surrounding Joyce wetlands. The canal also is a conduit for salt water during drought and storm surges, and has been a major contributor to the demise of the baldcypress – water tupelo forest in the southern half of the Joyce wetlands. The Mississippi River Gulf Outlet (MRGO), built in the early 1960's, also was a major contributor to saltwater intrusion and wetland degradation in the area (Shaffer et al., 2009b). The MRGO was closed in 2009 and salinities have significantly declined (Comite Resources et al., 2012).

During the same period I-55 was being constructed, South Slough was dredged, which directed flow from Selsers and Big Branch creeks, as well as diffuse runoff from the north, to the canal running parallel to I-55 (Fig. 1). The spoil generated from dredging of South Slough was placed on the south side of the canal, effectively blocking the flow of runoff from the upland watershed into the eastern Joyce wetlands and Four Mile Marsh. The 86-km^2 watershed generates an average of about 385,000 m^3 day^{-1} or about 30 times the effluent discharge, but all of this fresh water and associated nutrients and sediments ceased to flow into the Joyce wetlands once South Slough was constructed.

Since the construction of South Slough and the I-55 canal, saltwater intrusion has killed large areas of baldcypress – water tupelo forest in the Joyce wetlands. Surface water salinity of 3.5 ppt was measured immediately south of South Slough during summer, 2006, just prior to effluent discharge (Lundberg, 2008). Practically all baldcypress and water tupelo trees on Jones Island and south of Pass Manchac have been killed primarily due to high salinities (Shaffer et al., 2009a). The lower third of the Joyce wetlands also have experienced a high rate of forest loss due to high salinities. Water tupelo has been eliminated from the lower two-thirds of the Joyce wetlands due to salinity stress because tupelo is a strictly freshwater species while baldcypress can tolerate salinities of 3–4 ppt and short-term salinity increases of >5 ppt (Allen et al., 1994, 1996, 1997; Campo, 1996; Conner, 1994; Conner et al., 1997; McLeod et al., 1996; Shaffer et al., 2009a,b).

The Hammond Assimilation Wetland was divided into four sub-units for experimental purposes (Fig. 3). There also is a reference wetland located outside the influence of the discharge, located just north of South Slough (Fig. 3). Treated effluent is discharged into the treatment site and flows southward through the area. The 1.4 km discharge pipe has 900 outlets, but only 150–300 are opened at any given time. From an experimental standpoint, this enabled the flow to be concentrated in certain areas or, conversely, spread out across the entire system. To minimize disturbance to the marsh during system operation and data collection, four 200 m boardwalks were built equidistant and perpendicular to the outflow system.

Fig. 3. Map of Four Mile Marsh and the Hammond Assimilation Wetland. Shading indicates the location of experimental sub-units 1–4, the control site, and the discharge pipe.

3.2. Methods

3.2.1. Nutrient overloading

Surface water samples were collected during spring of 2007 along four transects running perpendicular from the outfall pipe, with samples taken every 50 m out to 300 m, and then every 100 m out to 700 m. In addition, long-term (2007–2013) surface water quality was collected quarterly at the outfall pipe, in the assimilation wetlands (Tmt), at a Mid site just south of the Four Mile Marsh at a boardwalk in the Joyce Wildlife Management Area, and where water exits the Joyce wetlands (Out). Water samples were taken in acid-washed polyethylene bottles, stored on ice, and taken to the laboratory for processing. Within 12-h, 60 ml from each water sample were filtered through pre-rinsed 25 mm 0.45 μm Whatman GF/F glass fiber filters into acid-washed bottles and frozen. Ammonium (NH_4–N) was determined by the automated phenate method, phosphate (PO_4–P) by the automated ascorbic acid reduction method, and silicon (SiO_4–Si) by the automated molybdate reagent/oxalic acid method (APHA, 2005). TN and TP were determined by methods described by Valderrama (1981).

3.2.2. Baldcypress biomass production

Eighty-eight seedlings were planted in groups of eight along each sub-unit. The seedlings were planted on 2.5 m centers every 50 m from the discharge pipe out to 300 m, then every 100 m out to 700 m. Each seedling was protected with a nutria-exclusion device and labeled with a unique metal identification tag. Trunk diameters were measured at 40 cm above the soil surface using digital calipers. Subsequent measurements were made at the end of the growing season (Lundberg et al., 2011).

To quantify biomass production of mature baldcypress located within 20 m of the outfall pipe, 22 trees were tagged and diameter growth was followed over the 2009 and 2010 growing seasons. We compare these growth rates with those from an 11-year study of over 1000 baldcypress in the largely declining Maurepas swamp (Shaffer et al., 2009a) and the nearby Joyce forest where we also followed growth of 22 edge and 22 interior trees.

3.2.3. Herbaceous biomass production

Eleven paired 4 m^2 permanent plots were established along each sub-unit at the same distances from outfall as the baldcypress seedlings. During August 2006 (prior to initiation of discharge), and spring and fall of 2007, two 0.25 m^2 clip plots were randomly removed from each paired plot. Samples were cold-stored until being dried and weighed.

3.2.4. Nutria herbivory

During the period when the rate of marsh deterioration was highest, we observed large areas of nutria eatouts, where all of the aboveground vegetation had been eaten to the wetland surface. We instituted a program of shooting to control nutria populations. During spring of 2008, to experimentally determine if the conversion from wetland to open water was caused primarily by nutria, ten 16-m^2 exclosures were constructed using 2-m wide vinyl-coated crab wire dug approximately 0.4 m into the ground. Ten 16-m^2 paired controls were demarked with PVC pipes. All of the exclosures and controls were planted with nine individuals of southern cattail (*Typha domingensis*).

As a second form of herbivore exclusion, 50 cm × 50 cm floating marsh 'pillows' were constructed out of vinyl-coated crab wire, and sprigs of maidencane (*Panicum hemitomon*) were enclosed within,

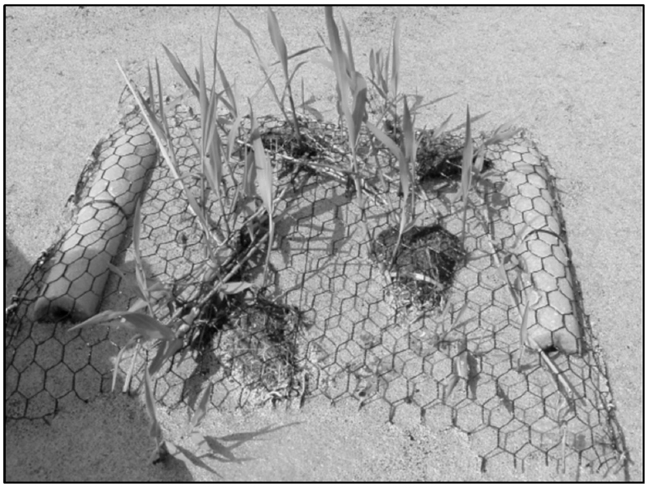

Fig. 4. Photograph of an herbivore exclusion 'marsh pillow' containing maidencane (*Panicum hemitomon*).

along with foam floatation (Fig. 4). Fifty of these pillows were placed along boardwalks in sub-units 1 and 4 (Fig. 3).

To quantify belowground plant biomass in the presence and absence of nutria, 50-cm deep, 9-cm diameter cores were extracted inside and outside of each 16-m^2 exclosure. Roots from the cores were carefully washed from the soil through wire mesh sieves. Live and dead belowground biomass was separated and live biomass dried at 65 °C and weighed (Valeila et al., 1976; Symbula and Day, 1988; Fitter, 2002).

3.2.5. Waterfowl herbivory

In an attempt to tease apart the herbivory impacts due to nutria and waterfowl, we constructed two 20-m × 40-m nutria exclosures during the spring and summer of 2010. While waterfowl cannot land in the 4 × 4 m exclosures, the larger cages enable waterfowl to forage, yet prevent entry by nutria (Shaffer et al., 1992).

3.2.6. Reduced belowground biomass

Belowground live biomass of bulltongue (*Sagittaria lancifolia*) was measured along the 700 m transects in sub-units 1 and 4, from the zone of discharge south toward the Joyce forest. We cored through marsh with this single species to test the effect of nutrients on belowground biomass at varying distances from the outfall source; bulltongue is the dominate if not only herbaceous species present from 0 m to 700 m from discharge.

From spring 2004 through fall 2008 we conducted a mesocosm experiment on the above- and belowground biomass production of eleven woody and herbaceous wetland plant species common to coastal Louisiana (Carrell, 2009; Hillmann, 2011). One-hundred-forty-four 200-liter vessels were planted with all eleven species and these were networked to 3000-l vessels that contained four water qualities (fresh with no fertilizer, fresh with time-released fertilizer, 3 ppt, and 6 ppt salinity) crossed with three hydrologies (mesic, permanent flooding, continuous circulation). At the end of the growing season of 2008, two 9-cm diameter, 30-cm long cores were extracted from the upper 30 cm of each vessel and two from the 30 cm to 60 cm depth (576 cores in all). Root material was carefully washed through a series of sieves and live roots were separated from dead, then dried and weighed for overall live belowground biomass across the eleven species (Valeila et al., 1976; Symbula and Day, 1988; Fitter, 2002).

3.2.7. Decomposition

Two hundred 10 cm × 15 cm bags sewn from fiberglass window screen were filled with 70 g of dried leaf litter from Manchac/Maurepas wetlands, weighed and tagged. The decomposition bags were deployed at 50, 100, 200, 400, and 600 m from the discharge system, along subunits 3 and 4. Bags were placed in 4 m × 4 m herbivore exclosure cages and open controls. Five pairs of bags were laced linearly together with braided fishing line. One of each pair was placed at the soil/water interface and the other buried approximately 12 cm deep. Samples were collected on a time series of 1, 2, 3, 6, and 12 months. During each collection, a bag from each orientation from the control and exclosure, at each distance, was collected from each of the two subunits. Bags were carefully rinsed, dried at 65 °C, and weighed.

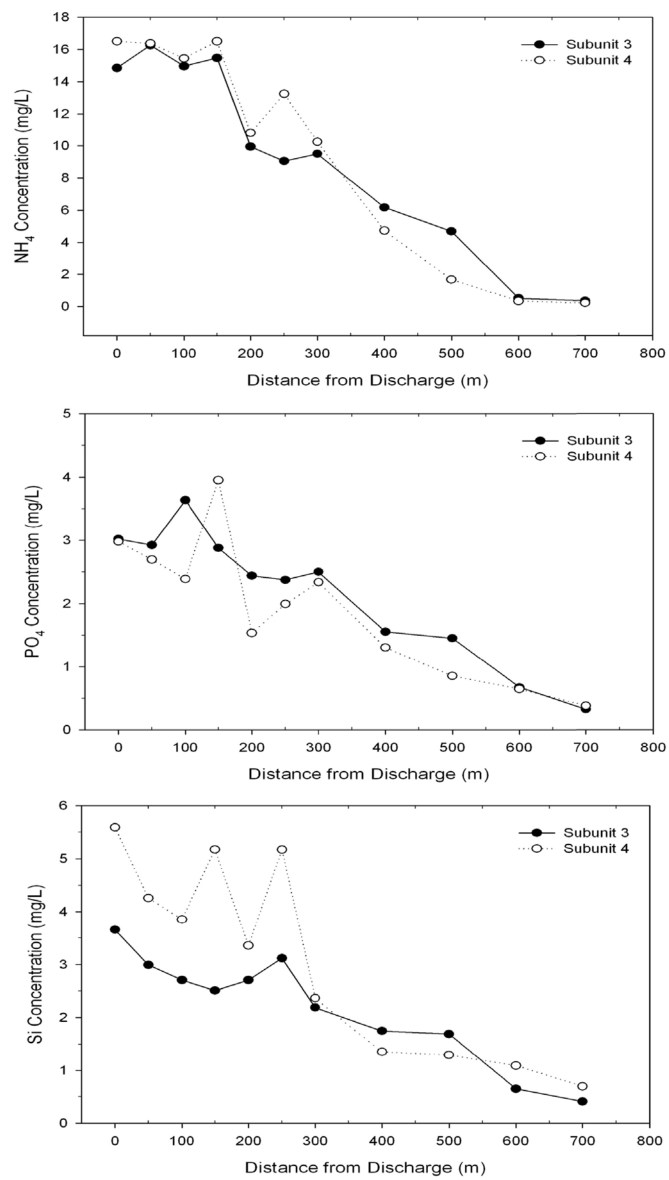

Fig. 5. Surface water quality (NH_4, PO_4, and Si) at various distances from the discharge pipe in the Hammond Assimilation Wetland (redrawn from Lundberg, 2008).

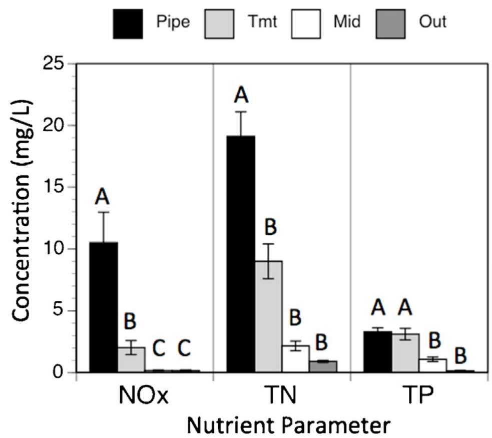

Fig. 6. Long-term (2007–2013) water quality data from the Hammond Assimilation Wetland (±1 s.e.). NO_x: nitrate + nitrite; TN: total nitrogen; TP: total phosphorus. Bars with different letters differ according to Tukey–Kramer multiple comparison.

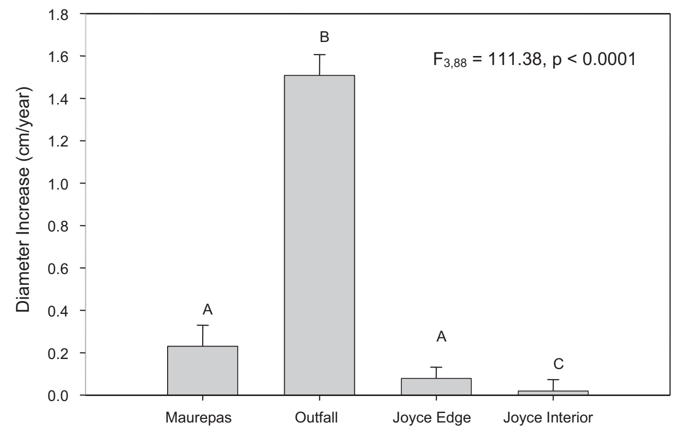

Fig. 8. Diameter increase of mature baldcypress trees growing in the Maurepas swamp, the outfall area of the Hammond Assimilation Wetland, the northern edge of the Joyce WMA, and the interior Joyce forest.

4. Results and discussion

4.1. Excessive nutrients

A linear decrease occurred in the concentrations of ammonium, phosphate and silicate from the outfall pipe along the 700 m transect. Inorganic nutrients were essentially non-detectable 600 m from the outfall pipe (Fig. 5). Long-term surface water quality data from 2007 to 2013 reveals substantial decreases in NO_x, TN, and TP as effluent passed through the Joyce wetlands (Fig. 6). Thus, the Hammond Assimilation Wetland is achieving one of its primary goals; that of reducing nutrient concentrations of overlying waters. Because nutrient concentrations reach background conditions (i.e., ≤3 mg/L TN and ≤1 mg/L TP; Hunter et al., 2009b) within 600 m of the discharge, the capacity of the wetland to reduce nitrogen and phosphorus concentrations has not been exceeded. Nutrient loading rates are calculated prior to any assimilation project to ensure that overloading does not occur (Day et al., 2004).

4.2. Baldcypress biomass production

Baldcypress seedling aboveground production followed a remarkably similar pattern as that of inorganic nutrients (Fig. 7). Growth was greatest at the outfall pipe and followed a linear decrease to 700 m from discharge (linear contrast $F_{1,218} = 69.71$, $p < 0.0001$).

The diameter increase of mature baldcypress trees located along the outfall pipe was five times greater than that of the Maurepas swamp and ten-fold higher then trees at Joyce (Fig. 8). We also planted hundreds of baldcypress seedlings within 20 m of the outfall system in 2008 that now average 8-m tall and are growing 2.01 cm y^{-1} (±0.08 cm y^{-1} S.E.) in diameter. There have been numerous studies showing either increased growth or no effect to baldcypress that are exposed to highly nutrified water. For example, Brantley et al. (2008) found significantly higher cypress growth downstream of effluent discharged from the Mandeville wastewater treatment plant. Shaffer et al. (2009b) found increased growth rates in the Maurepas basin in areas receiving regular non-point source inputs, as did Effler et al. (2006) for trees given nutrient amendments. Similar increases in growth also have been reported for other wetlands (Brown, 1981; Conner et al., 1981). Hunter et al. (2009a) found slightly higher, but not significant, cypress growth at the Breaux Bridge assimilation wetlands. Total NPP was highest at the treatment sites for the assimilation wetlands for the town of Amelia, LA (Day et al., 2006).

4.3. Herbaceous biomass production

Initially, the wetland vegetation responded vigorously to the new inputs of fresh water and nutrients, with a near doubling of

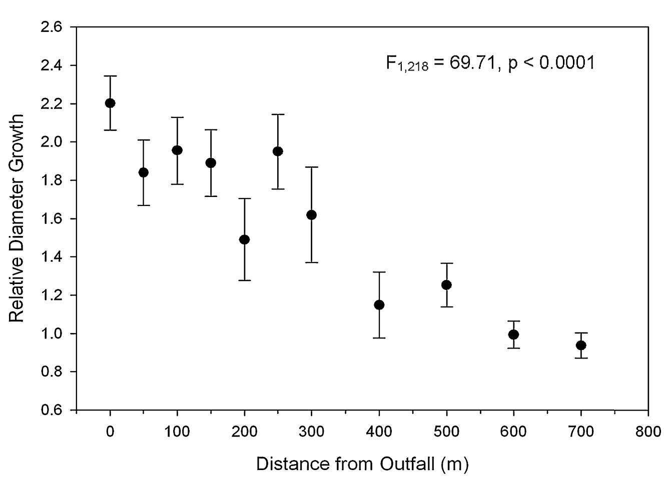

Fig. 7. Mean (±1 s.e.) relative basal diameter growth of baldcypress seedlings at various distances from the discharge pipe at experimental sub-units 1–4 (redrawn from Lundberg et al., 2011). Statistics are for linear contrast from 0 m to 700 m.

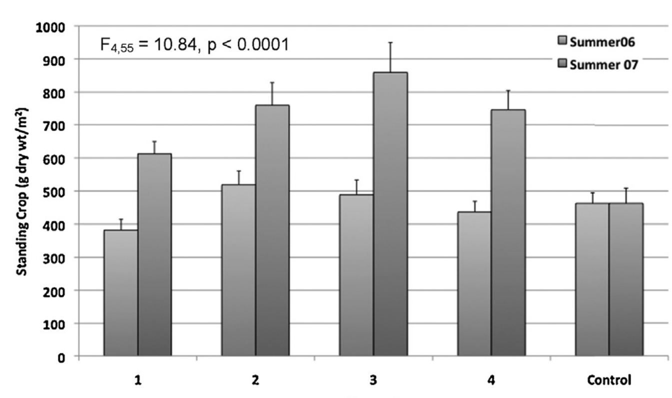

Fig. 9. Mean (±1 s.e.) aboveground herbaceous standing crop at the experimental sub-units and the control site (statistics are for 2007 growing season). These data were collected prior to marsh deterioration.

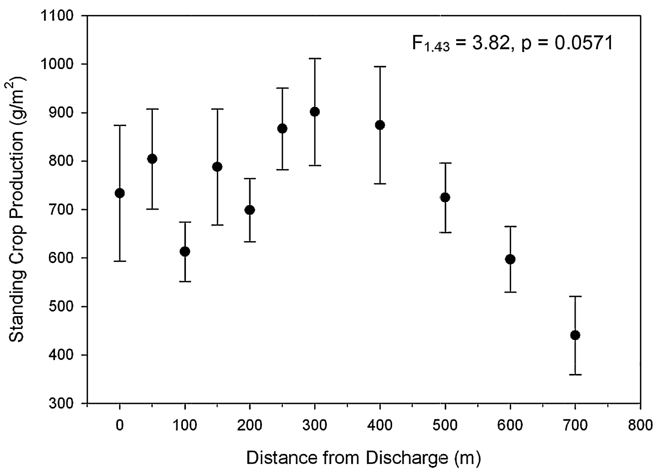

Fig. 10. Mean (±1 s.e.) aboveground herbaceous standing crop of permanent study plots at various distances from the discharge pipe (marginally significant linear contrast is from 400 m to 700 m).

biomass compared to controls (Fig. 9; 2007 growing season $F_{4,55} = 10.84$, $p < 0.0001$) and a clear individual sub-unit pattern of before (2006 growing season) vs. after discharge (2007 growing season). Herbaceous production was nearly constant to 400 m followed by a marginally significant linear decline (Fig. 10; $F_{1,43} = 3.82$, $p = 0.0571$), almost certainly due to nutrient limitation. However, by late fall 2007 the assimilation wetlands began to deteriorate, and within months nearly the entire area south of the discharge pipe had converted to open water or mudflat.

4.4. Nutria herbivory

Based on data from manipulative experiments as well as observations of nutria activity, it is clear that nutria were the dominant cause of marsh deterioration in the zone near the discharge pipe (Lundberg, 2008). After effluent discharge was initiated in November 2006, there was robust vegetative growth at the area receiving discharge, with greatly increased net primary production during the 2007 growing season (Fig. 9), attributable to the increased nutrient and freshwater input. Moreover, there was a linear decrease in herbaceous biomass production from 400–700 m from the outfall system (Fig. 10), indicating that nutrients were enhancing biomass near the outfall system.

By fall of 2007, large numbers of nutria were observed feeding on vegetation in the assimilation wetland. The animals were initially concentrated to the east and moved westward and southward in search of fresh vegetation. By the end of the 2007 winter, the wetland was mostly denuded.

In the nutria-exclosure experiment, *T. domingensis* displayed nearly 100% cover inside of all ten exclosures within a 3-month period. In stark contrast, cattail in all ten controls was completely destroyed within 48 h of planting. The control plots were replanted four times, and each time suffered 100% mortality due to nutria herbivory. Belowground biomass was nearly 3-fold higher inside of exclosures ($F_{1,16} = 30.04$, $p = 0.0003$) than in controls (Fig. 11). Furthermore, sub-unit 4 had higher belowground biomass than sub-unit 1 ($F_{1,16} = 19.52$, $p = 0.002$), and subunit 4 is where the effluent has been primarily concentrated (Fig. 12).

After concluding that nutria were a major detriment to the area, they were hunted aggressively in 2008–2009, with an estimated 2000 killed (Chris Carrell, unpublished data), and we have continued to shoot nutria since. Although both nutria and waterfowl foraged on the vegetation growing through the wire of the maidencane (*Panicum hemitomon*) floating marsh pillows, re-growth has occurred from roots inside the pillows.

4.5. Waterfowl herbivory

Shortly following initiation of effluent discharge, waterfowl recruited to the area *en masse*. Surveys indicate a 3-fold increase in waterfowl abundance at the Hammond Assimilation Wetland compared to other areas of the Manchac/Maurepas system (David Brown, Eastern Kentucky University, unpublished data). During winter months, approximately 2000 waterfowl use this area daily (Curtis Hymel, unpublished data). Within 4 months of constructing the 20-m × 40-m nutria exclusion cages, the caged areas nearly filled with herbaceous vegetation. However, by mid-January 2010, waterfowl had consumed nearly all vegetation in the cages except for giant cutgrass (*Zizaniopsis miliacea*). We now know that waterfowl, especially during the winter months, are currently eating as much or more herbaceous vegetation as are nutria. Our 16 m^2 exclosures, which prevent herbivory of both nutria and waterfowl, remain nearly 100% vegetated.

Over the past several years, the shallow open water areas of the marsh have become prime waterfowl habitat and one of the most important waterfowl hunting areas in the Pontchartrain Basin. Because the area is public, a part of the State Joyce Wildlife Management Area, it is open to all hunters. According to biologists at the Louisiana Department of Wildlife and Fisheries, nearly

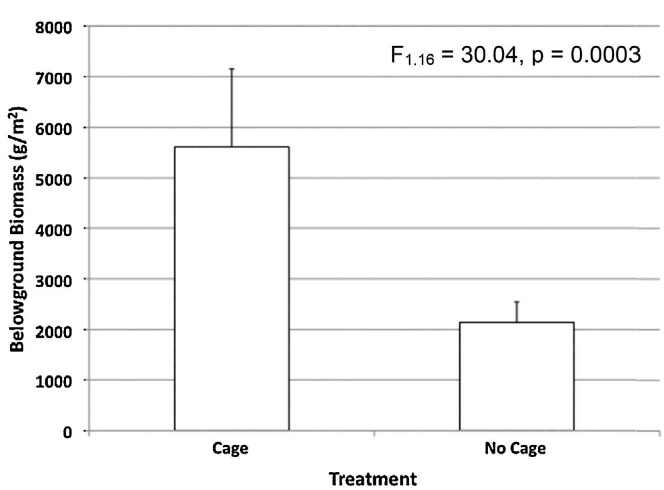

Fig. 11. Live belowground biomass measured within nutria exclosures (cages) compared to unprotected nearby sites.

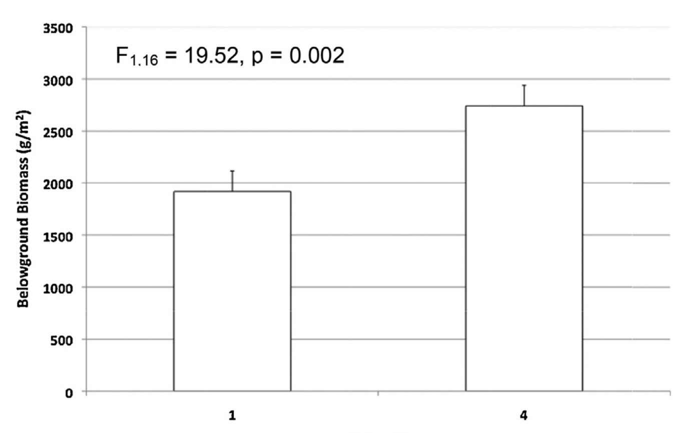

Fig. 12. Belowground biomass of bulltongue (*S. lancifolia*) at sub-unit 1 (lowest nutrient loading) and sub-unit 4 (highest nutrient loading).

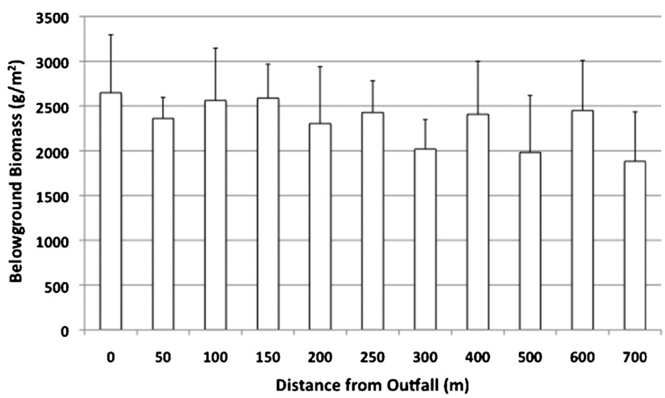

Fig. 13. Belowground biomass of bulltongue (*S. lancifolia*) with distance from the discharge pipe showing no significant trend.

1000 waterfowl were taken from the area during the 2013–2014 season (Christian Winslow, La. Dept of Wildlife and Fisheries). Biologists in the Department want to preserve some of the shallow open water area to continue to provide habitat for waterfowl.

4.6. Reduced belowground biomass

Live belowground biomass of bulltongue averaged 2330 g dry wt m^{-2} and ranged between 286 and 5676 g dry wt m^{-2} with no trend of increasing or decreasing biomass from 0 m to 700 m of the outfall system (Fig. 13).

In the mesocosm experiment, root to shoot ratio decreased by about 50% in the fertilized treatment, as expected (Hillmann, 2011). However, the total amount of belowground biomass was about double that of the other three water quality treatments (fresh with no fertilizer, 3 ppt, 6 ppt), especially in the 30- to 60-cm deep cores ($F_{3,135} = 16.44$, $p < 0.001$). Both aboveground and belowground biomass responded positively to fertilization, but the aboveground response was far greater than the belowground response (Hillmann, 2011). Moreover, organic accretion in the fertilized treatments averaged a 2 cm yr^{-1} elevation gain compared to about 0.4 cm yr^{-1} loss in the other water quality treatments.

It is unlikely that excessive nutrient loading led directly to the marsh deterioration. Nutrient loading to the marsh area was low to moderate (15.7 g N m^{-2} yr^{-1} and 1.6 g P m^{-2} yr^{-1}). There is no example that we know of where similar nutrient loading led to marsh deterioration, especially in such a short period of time. Coastal marshes have received much higher loading for long periods of time without converting to open water. For example, nitrogen and phosphorus has been applied to the Great Sippewissett salt marsh in Massachusetts since the early 1970s, with total nitrogen loading rates ranging from 18 to greater than 157 g N m^{-2} yr^{-1}, with no major wetland deterioration (Valiela et al., 1975, 1976; Giblin et al., 1980, 1986; Turner et al., 2009; Fox et al., 2012). Turner et al. (2009) reported that there was a reduction of belowground biomass, however, the Sippewissett salt marsh remains intact after more than 35 years of nutrient additions. Fox et al. (2012) suggested that the accretion reduction might be overstated because of solubility of ash. Anisfeld and Hill (2012) carried out a 5-year study of fertilization on a Connecticut salt marsh. They concluded that neither nitrogen nor phosphorus fertilization led to elevation loss, reduced soil carbon, or a decrease in belowground primary production. They disagreed with the findings of Darby and Turner (2008b). They stated "First, Darby and Turner (2008b) arbitrarily exclude one out of two *S. alterniflora* experiments (the "low fertilization" experiment) as being an outlier. The excluded experiment showed higher, rather than lower, biomass in fertilized plots, although the difference was not significant. Second, closer examination of the one *S. alterniflora* experiment selected by Darby and Turner (2008b; the "high fertilization" experiment) shows that fertilized and control plots were not statistically different in that experiment either. Third, Darby and Turner (2008b) examine only the August data rather than the entire dataset. Analysis of increases and decreases in both live and dead material over the entire season (as done by Valiela et al. (1976) in their Table 2) shows that fertilization leads to either large increases in estimated belowground production (one out of two *S. alterniflora* experiments, both *Spartina patens* experiments) or no change (the other *S. alterniflora* experiment). Finally, Valiela et al. (1976) also measured *S. alterniflora* belowground productivity using ingrowth cores and found that fertilization did not decrease belowground production but rather increased it."

Likewise, shorter-term nutrient additions to Louisiana salt marshes that resulted in reduced belowground biomass and soil strength had over an order of magnitude higher loading rates (225 g N m^{-2} yr^{-1}) than at the Hammond site (Darby and Turner, 2008a,b,c; Turner, 2010). Moreover, the fertilizer that Darby and Turner (2008a,b,c) used contained 425 g S m^{-2} (Nyman, 2014), and in wetlands sulfur rapidly reduces to highly toxic sulfides. In addition, vegetative cover at the Hammond Assimilation Wetland is highest closest to the outfall area and growth of mature baldcypress near the discharge area is over five-fold higher than that of the Maurepas swamp and ten-fold higher than that of the nearby Joyce forest. Finally, Morris et al. (2013a,b) found that eight years of fertilization in a *S. alterniflora* salt marsh in North Inlet, SC, caused higher aboveground biomass which resulted in more friction that in turn caused mineral sediment to fall out of the water column resulting in increased elevations, compared to the control marsh. A review of the literature shows that there are many more studies that show positive impacts of added nutrients on marshes (Haines and Dunn, 1976; Valiela et al., 1976; Buresh et al., 1980; Haines, 1979; Stevenson and Day, 1996; Shipley and Meziane, 2002; Day et al., 2004, 2006; Ravit et al., 2007; Hunter et al., 2009a, b; Carrell, 2009; Shaffer et al., 2009a; Hillmann, 2011; Priest, 2011; Zhang et al., 2013; Morris et al., 2013a) than negative ones (Morris and Bradley, 1999; Darby and Turner, 2008a,b,c; Swarzinsky et al., 2008; Wigand et al., 2009; Deegan et al., 2012).

There are a number of coastal marsh systems in Louisiana where long-term nutrient loading has not caused marsh deterioration. The Atchafalaya and Wax Lake deltas have developed with very high river input, and the region affected by Atchafalaya River input has low wetland loss rates compared to most of the coastal zone (Britsch and Dunbar, 1993; Barras et al., 2008; Couvillion et al., 2011). Day et al. (2011a) reported that salt marshes near lower Fourleague Bay with strong riverine input have been stable for over a half century while salt marshes east of Bayou Terrebonne, with no river influence, have high rates of deterioration. Neither of these sites have significant hydrological alteration. Day et al. (2011b) reported similar findings for the northwestern Mediterranean where coastal marshes with riverine input had much higher rates of accretion and elevation gain and belowground biomass and were likely to survive accelerated sea-level rise. Even in the Po River Delta which has nitrate concentrations near 10 mg/l and little suspended sediments, accretion at the river mouth sites was 3–4 times higher than at non-riverine influenced sites.

In contrast, Swarzenski et al. (2008) reported that floating *P. hemitomon* marshes in the Bayou Penchant region affected by inflow of Atchafalaya were in a state of deterioration with the organic matter substrate more decomposed and higher levels of sulfur than areas that do not receive river water. They concluded that the continual input of river water leads to a reducing soil environment, increased sulfide and inorganic nutrients in

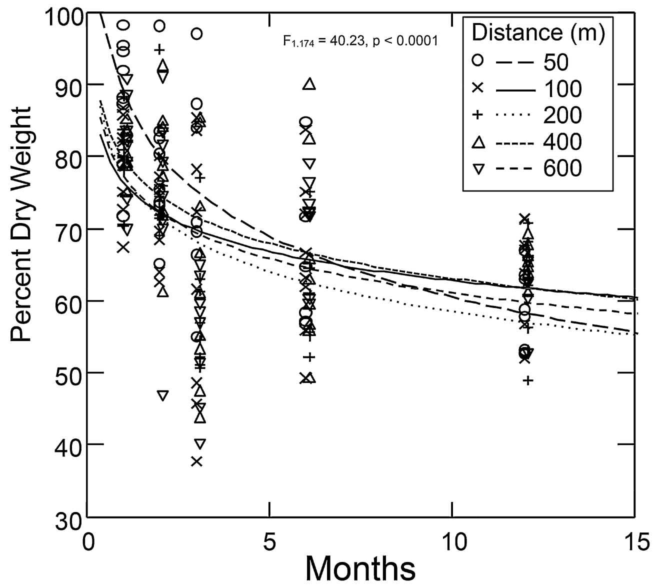

Fig. 14. Percent dry matter remaining across the 12-month decomposition study. One exponential decay curve is shown for each distance from the discharge system to show that rates of decomposition did not differ at the varying distances from outfall ($F_{4,174}$ = 1.45, p = 0.220) even though overall exponential decay curve is highly significant ($p < 0.0001$).

porewater, and internally generated alkalinity, and that these conditions lead to organic matter decomposition and root decomposition. However, Swarzenski et al. (2008) did not consider the impact of grazing by nutria. Sasser et al. (2004) conducted a study in the same Bayou Penchant area on the impact of excluding nutria on floating marsh. They found that excluding nutria resulted in a dramatic increase in both aboveground and belowground growth. The results of Swarzenski et al. (2008) and Sasser et al. (2004) suggest that there is a complex interaction among freshwater input, nutrients, and nutria grazing. This topic needs to be studied in more detail before definitive conclusions can be drawn. Recently, it has been demonstrated that nutria prefer fertilized wetland herbaceous plants to unfertilized ones (Ialeggio and Nyman, 2014).

Izdepski et al. (2009) reported that discharge of treated effluent into a shallow open water area near Thibodaux, LA, led to the formation of a freshwater floating marsh. In the Central Wetlands Unit (CWU) in St. Bernard Parish, the only areas where baldcypress and low salinity vegetation survived saltwater intrusion from the MRGO were locations where surface runoff was pumped into the wetlands (Shaffer et al., 2009b). At the Gore pumping station, where surface runoff combined with treated effluent has been pumped into wetlands of the CWU since the early 1960s, a relic baldcypress forest has survived and soil strength is relatively high compared to the rest of the CWU.

Hurricane Katrina caused the loss of 212 km^2 of wetlands primarily in the Breton Sound estuary, which is influenced by the Caernarvon river diversion (Barras, 2006). Howes et al. (2010) reported that soils of low salinity wetlands in the upper Breton Sound estuary had significantly lower shear strength compared to higher salinity wetlands. They concluded that this was due to biomorphological differences between high and low salinity plant species, which may be exacerbated by low mineral sediment and high freshwater and nutrient inputs. Kearney et al. (2011) analyzed wetland changes at Caernarvon before and after the two hurricanes. They concluded that the "vulnerability to storm damage reflects the introduction of nutrients in the freshwater diversions, . . . which promotes poor rhizome and root growth in marshes where belowground biomass historically played the dominant role in vertical accretion." However, Kearney et al. (2011) provided no nutrient data to show elevated levels. Day et al. (2013) reviewed a number of studies on the influence of the Caernarvon diversion on wetlands and waters of the Breton Sound estuary. They reported that nutrients in diverted water flowing over marshes, especially nitrate, decreased rapidly and nitrate and suspended sediment concentrations in interior marshes were near non-detectable. Day et al. (2009, 2013), measured above- and belowground biomass and decomposition at streamside marshes in near, intermediate, and far locations from the Caernarvon diversion and at a reference location. Belowground biomass was high and there were little to no differences in decomposition among sites. Water level analysis at Caernarvon showed that percent time flooding increased from less than 20% in the late 1990s to between 60% and 90% from 2001 to 2010 (Gregg Snedden, USGS, personal communication). Day et al. (2011a) reported that salt marshes that were flooded about 15% of the time were well-drained and healthy while marshes flooded 85% of the time deteriorated. Shaffer et al. (1992) found that wetland vegetation experiencing greater than 50% flooding converted to mudflats in the Atchafalaya. Because nutrients are confounded with the floodwaters that carry them, we believe that the negative effects of excessive flooding are being inaccurately attributed to excessive nutrient loading.

4.7. Nutrients impacting decomposition

Though highly variable within each time period, decomposition followed the traditional exponential decay curve over the 12-month period ($F_{1,174}$ = 40.23, $p < 0.0001$, R^2 = 0.231), but rates of decrease did not differ across distance from outfall ($F_{4,174}$ = 1.45, p = 0.220; Fig. 14). No difference in decomposition occurred for herbivore exclosures vs. open areas, or for the surface vs. buried bag orientation.

4.8. Other hypotheses for the conversion of marsh to open water

The following hypotheses have been raised concerning the conversion of wetlands to open water in the Hammond Assimilation Wetland. Below we have listed those hypotheses along with discussion, supported by data when possible.

4.8.1. pH impacting decomposition

The pH of the effluent was higher than the pH of the surface water in the assimilation wetland, and this higher pH increased decomposition rates and impacted the organic matter content and strength of soil while it is correct that mean pH in the assimilation wetland was lower prior to discharge (about 5.5–6) than after discharge began (about 7), we know of no studies that have determined how such an increase in pH would correlate to an increase in decomposition rates. Organic matter decomposition in wetlands is regulated by a number of factors, including dissolved oxygen, temperature, pH, organic matter quality, nutrients, and the availability of other terminal electron acceptors (Craft, 2001; Mitsch and Gosselink, 2007). In addition, both bacteria and fungi contribute to decomposition, and optimum pH for bacterial decomposition occurs between 6-8, while optimum pH for fungal decomposition occurs between 4 and 6. Higher rates of decomposition would be expected after grazing by nutria, as they are extremely inefficient herbivores that leave most of the clipped vegetation to decompose. Similar to the nutrient loading pattern, pH would reach background levels at some distance from the discharge system. Therefore, if an increase in pH were to increase decomposition rates, then decomposition rates would be higher nearest the discharge; they are not (Fig. 14).

4.8.2. Disease

4.8.2.1. Plant diseases induced by high nutrients caused vegetation death at the Hammond Assimilation Wetland. Photographs have been presented showing indications of various plant diseases (e.g., *Fusarium* fungi) at the assimilation wetland, but no quantitative data have been presented to show whether these were a common occurrence in the marsh or whether they have been reported to affect marsh growth. There have been no data presented on any studies on disease impacts in marshes. Until such information is presented, this should be considered as an untested hypothesis. It is known that fertilized agricultural crops are more susceptible to diseases but very little is known about wetland vegetation. It might be reasonable to think that disease could affect individual species; however, it is extremely unlikely that a pathogen or pathogens could impact an entire wetland of diverse plant species because each species would vary in its susceptibility to the pathogen (Dr. Reymond Schneider and Dr. Laurence Datnoff, LSU Plant Pathology Department, personal communication). This is a topic area that should be investigated in more detail.

4.8.3. Toxic effluent

4.8.3.1. There is some chemical (e.g., chlorine) in the effluent that caused the vegetation dieback. Stem diameter growth of the 440 baldcypress seedlings planted perpendicular to the discharge system was highest near the outfall and decreased linearly with distance from discharge pipe (Fig. 8). Marsh regrowth was most vigorous and persistent near the discharge. Any toxicity effect should be manifested close to the discharge where any toxic material concentrations would be highest. Interestingly, inorganic nutrient concentrations followed the same pattern as baldcypress seedling production with distance from outfall to the Joyce forest (Fig. 5). Concentrations of ammonia, phosphorus, and silica were reduced to very low levels after water passed through the assimilation wetland. In addition, acute toxicity testing and analysis for metals, volatile organic compounds, and other pollutants is required on all municipal discharges, not just those going into assimilation wetlands. Effluent going into the Hammond wetland did exceed LPDES permit limits for copper and zinc due to old piping but the city is working to correct this problem and no other metals have recently exceeded permit limits in the effluent during repeated testing.

4.8.4. Flood stress

4.8.4.1. Excessive inundation from the discharge of effluent caused the vegetation dieback. In the outfall area, flood levels have increased by approximately 30 cm since initiation of discharge in fall, 2006, although they decrease to background levels near the southern end of the marsh. As of May 2011, two water control structures with flap gates were installed at the northeastern and northwestern corners of the assimilation wetland to allow water to drain into South Slough. This was done to allow water on one side of the assimilation wetland to be lowered when discharge is at the other side (a distance of 1.4 km). Alternatively, discharge can be concentrated at the center of the system and both culverts opened to maximize drawdown potential; under this scenario water still flows over at least 500 m of wetland for adequate bioremediation. However, to date water levels have remained excessive and we are now suggesting that a new outfall system be constructed on the west side of I-55 creating two independent systems. This would enable pulsing of the effluent for several months to one assimilation wetland, allowing complete drawdown of the other wetland. At present, plans are being developed for the new outfall system.

5. Conclusions

Based on data from manipulative mesocosm and field experiments as well as observations of nutria eatouts, it is clear that nutria were the dominant cause of vegetation loss at the Hammond Assimilation Wetland. This is supported by studies showing that when nutria were excluded, the marsh flourished, local extinction of *T. domingensis*, except in exclosures (a nitrophilic species that generally dominates under eutrophied conditions, but is a preferred food of nutria), and recovery of the marsh following aggressive nutria control (especially within 200 m of the discharge area where water levels are highest). However, the marsh deterioration due to nutria led to a cascade of additional impacts. For example, when the marsh opened up, large numbers of waterfowl were attracted to the ponded areas. Grazing of herbaceous vegetation by waterfowl is as important as nutria during winter months. Because of the shallow ponds, the area has become a prime habitat for waterfowl. As restoration proceeds, an important question is: Is it desirable to maintain some open water for waterfowl habitat, or try to restore the whole area back to emergent wetlands?

It is unlikely that excessive nutrient loading led directly to the marsh deterioration because, as discussed above, nutrient loading to the marsh area was low to moderate. There is no example that we know of where similar nutrient loading led to marsh deterioration and conversion to open water in such a short period of time (less than a year). Fertilization experiments that have lasted for decades have shown the opposite result. There are also a number of coastal marsh systems in Louisiana where long-term nutrient loading has not caused marsh deterioration, with the Atchafalaya region being the most notable. The land loss rate in the wetlands affected by the Atchafalaya River is the lowest in coastal Louisiana (Day et al., 2000; Barras et al., 2008; Couvillion et al., 2011). Day et al. (2011a) reported that salt marshes southeast of the Atchafalaya River have been stable for over a half-century, while salt marshes further east not in contact with river water have high rates of land loss. Day et al. (2011b) reported similar findings for the northwestern Mediterranean where coastal marshes with riverine input had much higher rates of accretion and elevation gain and were likely to survive accelerated sea-level rise.

We are currently in the planning stages for a pulsing system where the effluent can be discharged east and west of I-55. This would have the benefit of maintaining low salinities on both sides of the highway and an ideal pulsing paradigm could be established to optimize the hydrologic regime. Without question, the Hammond Assimilation Wetland has demonstrated that all assimilation wetlands in coastal Louisiana should be built so that water levels can be controlled and that, in herbaceous wetlands receiving effluent discharge, nutria control should be a crucial part of the management plan.

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