

Phosphorus Fluxes in a Restored Carolina Bay Wetland Following Eight Years of Restoration

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33 **ABSTRACT**

34 Restoring wetlands on agricultural land can release soil P to surface waters. Phosphorus is a
35 limiting nutrient in many freshwater systems, thus restricting its release will improve surface
36 water quality. A P balance was used to examine how P was cycling in a Carolina Bay wetland
37 eight years after restoration from prior-drained agricultural land. The change in soil P was
38 evaluated between archived samples taken at restoration (2005), and eight years after restoration
39 (2013). Measured P fluxes included atmospheric deposition, plant uptake, and loss to surface
40 water outflow. The soil total P pool at the time of restoration was 810 kg P ha⁻¹. No significant
41 ($\alpha=0.05$) decrease in the soil P pool was observed. Atmospheric deposition contributed 7 kg P ha⁻¹
42 ¹, plants accumulated 28 kg P ha⁻¹ and incorporated 27 P ha⁻¹ into woody biomass and 1 kg P ha⁻¹
43 on the forest floor litter, and 1.7 kg P ha⁻¹ was lost to surface waters draining the wetland.
44 Because the loss of P to surface waters was small, and because runoff water concentrations of P
45 declined through this period of study to concentrations below those likely to cause eutrophication
46 (< 0.1 mg L⁻¹), we concluded that the wetland was not contributing to the degradation of surface
47 water quality of nearby streams following restoration. Further, relatively “isolated” wetlands
48 such as that studied may be promising sites for future wetland mitigation projects due to limited
49 impacts on surface water quality.

50 **KEY WORDS**

51 Wetland restoration; water quality; isolated wetlands; converted wetlands; phosphorus

52 **INTRODUCTION**

53 Over 50% of the original wetlands in the conterminous 48 states of the U.S. were drained
54 primarily for food production between 1780 and 1980 (Dahl and Allord 1996). Since 1977,
55 federal and state programs have been enacted to reverse the loss by restoring drained areas to
56 their original wetland condition, frequently by plugging or filling drainage ditches. Wetland
57 restoration is accomplished, in part, to improve water quality. However, in cases where wetlands
58 are restored from agricultural land that is high in P from years of fertilization, saturated and
59 reduced soil conditions cause P to be released from the newly flooded wetland contributing to
60 eutrophication of nearby surface waters. Between 1997 and 2001 there was an estimated annual
61 net gain of 13,400 ha of wetlands nationally due to restoration of agricultural fields, while
62 between 2001 and 2003 the annual net gain more than doubled from previous periods (USDA-
63 NRCS 2013). Based on estimates from the North Carolina Division of Mitigation Services,
64 approximately two-thirds of restored wetlands in North Carolina originated from drained and
65 fertilized agricultural lands – equivalent to approximately 1500 ha since 1999 (Smith 2011).
66 Given that wetland restoration is increasing in the U.S., in part to improve water quality, it is
67 critical to determine whether restoration will actually contribute to pollution of P-sensitive
68 watersheds.

69 The problem of P dissolution in wetlands restored on agricultural fields has been observed
70 around the world. Recent studies of P dissolution have been done on peat soils in the Netherlands
71 (Van Dijk et al. 2004), soils used for dairy production in Florida (Pant and Reddy 2003), restored
72 lake fringe in Oregon (Aldous et al. 2007; Duff et al. 2009), and agricultural soils in the North

73 Carolina Coastal Plain (Ardon et al. 2010), and a Carolina Bay complex in North Carolina
74 (Bruland et al. 2003). These studies have shown that P dissolution is largely driven by Fe
75 reduction processes (Reddy and DeLaune 2008), along with other mechanisms including ligand
76 exchange (Earl et al. 1979; Lopez-Hernandez et al. 1986; Violante et al. 1991; Gerke 1992), P
77 mineralization from drying and rewetting cycles (Song et al. 2007), changes in pH (Jackson
78 1964; Ponnampereuma 1972; Stumm and Morgan 1981), and increased P diffusion (Turner and
79 Gilliam 1974a; Turner and Gilliam 1974b).

80 To ensure that wetland restoration and management practices do not contribute to pollution of
81 nutrient sensitive streams, a better understanding of P fluxes within and out of wetlands restored
82 from agricultural land is greatly needed in order to identify potential management strategies that
83 will reduce P loss. This study focused on a previously cultivated Carolina Bay wetland, known
84 as Juniper Bay, that was restored as a wetland and then monitored for P in drainage waters in
85 subsequent years (Vepraskas et al. 2010; Moorberg et al. 2014; Moorberg et al. 2017). The
86 objectives of this study were to estimate a P budget for a restored wetland by determining P
87 fluxes for soil storage, plant uptake, atmospheric input, and loss from drainage water.

88 **MATERIALS AND METHODS**

89 Juniper Bay is located in Robeson County, NC approximately 10 km south of Lumberton, NC
90 (34°30'30"N 79°01'30"W). In 1999, the North Carolina Department of Transportation (NCDOT)
91 purchased this drained Carolina Bay wetland to mitigate the destruction of nearby wetlands
92 caused by highway construction (Ewing 2003). The Bay is oval-shaped, oriented lengthwise
93 along a northwest-southeast transect, and is virtually flat with an area of 291 ha. Soils in Juniper
94 Bay include approximately 60% mineral soils (Leon sand; sandy, siliceous, thermic Aeric
95 Alaquods, USDA Soil Taxonomy) primarily at the edges (Figure 1, SC and SS mapping units),

96 with organic soils (Ponzer muck; loamy, mixed, dysic, thermic Terric Haplosaprists, USDA Soil
97 Taxonomy), occupying the remainder at the center (Figure 1, OC and OS mapping units). This
98 Bay was drained for agriculture beginning in 1971 by excavating a perimeter ditch around the
99 edge of the Bay, and installing primary and secondary ditches within the Bay to facilitate
100 drainage into a single surface water outlet on the southwestern edge of the wetland (Vepraskas et
101 al. 2005). Juniper Bay was fertilized and limed annually to meet soil-test recommendations. It
102 remained in crop production until 2001. Preliminary restoration efforts began in June 2003, and
103 wetland hydrology was restored in 2005 by filling primary ditches and plugging tertiary ditches,
104 leaving only the perimeter ditch intact. That perimeter ditch drains into one outlet on the
105 southwest side of the Bay.

106 A P-balance was developed to better understand the nature and relationships of P fluxes into and
107 out of Juniper Bay following restoration. The proposed P-balance in simplest form is shown in
108 Equation 1:

$$\Delta P_{\text{soil}} = P_{\text{inputs}} - P_{\text{outputs}} \pm E \quad [1]$$

109 where ΔP_{soil} is the change to the soil's total P (TP) pool to a depth of 1 m since restoration (2005-
110 2013). The P_{inputs} are sources of "new" P, while P_{outputs} include all mechanisms that remove P
111 from the soil. The E is an error term that accounts for errors in the determination of the change of
112 P in the soil, errors in P fluxes, and/or error or fluxes that have gone unrecognized.

113 We hypothesized that atmospheric deposition (P_{ATM}) is a major mechanism adding new P into
114 the Bay, but groundwater inflow (P_{GI}) could also be contributing P. Major ways for P to be
115 removed from the soils in Juniper Bay include: plant uptake (P_{PL}), groundwater outflow (P_{GO})
116 and surface water outflow (P_{SO}). Previous work by Pati (2006) showed that the perimeter ditch

117 would intercept most groundwater inflow into the Bay, thus transforming P_{GI} into P_{SO} which
 118 would drain out of the Bay through the outflow structure. In addition, Pati (2006) showed that
 119 the groundwater outflow component would be intercepted by the perimeter ditch as well, as long
 120 as the water levels in the ditch were managed to stay below a critical elevation. Such
 121 management of the perimeter ditch is currently practiced so that groundwater outflow from the
 122 Bay should be small. Huffman et al. (2007) estimated the net flow of ground and surface water
 123 into the Bay from the surrounding landscape was equivalent to 125 mm during the wet months of
 124 2004, with inflows entering the perimeter ditch on the NW, NE, and SE sides of the Bay, and
 125 groundwater outflows exiting on the SW side of the Bay. The impact of the perimeter ditch is
 126 such that the terms for P_{GO} , P_{SO} , and P_{GI} were combined with the assumption that contributions
 127 from groundwater inflow are minimal, and that surface water outflow is primarily from drainage
 128 from Juniper Bay. This assumption was tested and validated, as described in the supplementary
 129 material (SI Table 1 and SI Table 2; SI Figure 1, SI Figure 2, SI Figure 3, and SI Figure 4)

130 The P balance for Juniper Bay can be written with the defined inputs and outputs as:

$$\Delta P_{soil} = P_{ATM} - P_{PL} - [P_{GO} + P_{SO} - P_{GO}] \quad [2]$$

131 For simplicity, we combined the components P_{GO} , P_{SO} and P_{GI} into one term called $P_{OUTFLOW}$
 132 which was measured collectively at the outflow structure. The modified P balance used for this
 133 study is:

$$\Delta P_{soil} = P_{ATM} - P_{PL} - P_{OUTFLOW} \pm E \quad [3]$$

134 The volume of soil considered for Juniper Bay has a horizontal area defined by the perimeter
 135 ditch (Figure 1B) with the soil depth starting at the soil surface and extending to a depth of 1 m.

136 The 1 m depth was selected because previous work showed that the P increases from agricultural
137 applications in Juniper Bay were not observed below 1 m (Ewing 2003).

138 The change in the soil P pool was determined by measuring total phosphorus (TP_{soil}) in archived
139 soil samples (2005) and comparing those values with TP_{soil} found in present day samples
140 extracted from the same locations. The TP_{soil} concentrations for time-zero (2005) were
141 determined from two groups of archived soil samples. Prior to restoration, soils were sampled in
142 2000 from 48 soil pit locations to a depth of 1 m (Figure 1B) by Ewing et al. (2012), and in 2004
143 on a grid of 700 locations across the Bay to depths of 0-0.15 m and 0.15-0.3 m using soil push
144 probe. It was assumed that P concentrations in the subsoil (0.15-0.3 m) had not changed between
145 2000 and restoration in 2005 based on the observations made by Ewing et al. (2012) which noted
146 that subsoil P concentrations were low and did not exceed those found in nearby reference
147 Carolina Bay wetlands. Since Juniper Bay remained artificially drained from 2000 to 2004, soil P
148 should have remained immobile prior to restoration.

149 For each archived soil sample location, GPS coordinates were recorded in 2005 at the time of
150 sampling, thus allowing new samples to be extracted from the same locations. For the 0-0.15 m
151 and 0.15-0.3 depths 138 locations of the 700 total were selected for re-sampling and analysis
152 using an area-weighted, stratified random sampling scheme (Figure 1B). The samples were
153 separated into four strata based on the soil mapping units developed for restoration of Juniper
154 Bay during the NCDOT soil survey. All archived surface and subsoil sites were located using
155 GPS receivers with a wide area augmentation system correction and 2-5 m accuracy.

156 The study by Ewing et al. (2012) included 48 soil pit locations. These locations consisted of 24
157 pairs of pits - one at the crest (middle) of the fieldlet, and one adjacent to the ditch. They

158 observed large amounts of disturbance in the ditch pits due to maintenance and dredging of the
159 drainage ditches during agricultural production; therefore, only the crest pits were used in this
160 study. Also, Ewing et al. (2012) studied five pits from soils with histic epipedons at the transition
161 from mineral to organic soils. Because these histic soils represent a small area of the Bay, they
162 were also omitted from this study. The remaining 19 soil pits used in this study are shown in
163 Figure 1B. The 2013 soil samples were extracted using a soil auger for all three depths.

164 All soil samples were submitted to the North Carolina Department of Agriculture Soil Testing
165 Service for analysis of extractable P by the Mehlich-3 method (Mehlich 1984). Soil TP analysis
166 was performed on 25% of the re-sampled 0-15 cm and 15-30 cm depth soil samples that were
167 selected at random within each stratum (Figure 1B). Soil TP analysis was also performed on four
168 representative horizons from each pit location (Figure 1B). Soil TP was determined by nitric-
169 perchloric acid digestion (Carter 1993). The TP_{soil} determined on a mass per mass basis were re-
170 expressed as mass P per volume of soil for inclusion into the P-balance. Bulk densities reported
171 by Ewing et al. (2012) for the pre-restoration samples at all depths were used to convert the mass
172 of soil to its equivalent volume. Bulk density was determined again for the 2013 samples for the
173 0-15 and 15-30 cm depths using the core method (Grossman and Reinsch 2002). Samples were
174 collected in triplicate at each of the 19 soil pit locations and averaged across the four soil
175 mapping units for the 0-15 and 15-30 cm depths. Bulk densities for the 0.3 to 1.0 m depths were
176 assumed to be the same as the pre-restoration values as reported by Ewing et al. (2012).

177 Atmospheric deposition of P was monitored from May 2012 through June 2013 at three locations
178 within the wetland (Figure 1A), as described by Kreiser (2003). Samplers were installed adjacent
179 to existing rain gauges using a bulk rain water collection apparatus (SI Figure 5) modeled after
180 Likens et al. (1967) and Johnson and Swank (1973). Samples were collected every two to four

181 weeks and acidified for preservation. Samples were submitted to the North Carolina State
182 University Environmental and Agricultural Testing Service (Raleigh, North Carolina, USA) for
183 determination of dissolved reactive P (DRP) and dissolved total P (DTP). The average
184 concentration of DTP for this time period, along with historic rainfall data collected on site were
185 used to estimate P_{atm} from 2005 to 2013 over the entire area of the wetland.

186 Phosphorus uptake and accumulation by trees was estimated for the entire area of Juniper Bay.
187 The North Carolina Department of Environment and Natural Resources (now the North Carolina
188 Department of Environmental Quality, NCDEQ) planted wetland tree saplings throughout
189 Juniper Bay at the time of restoration (NCDEQ 2010). Between 2005 and 2010, NCDEQ (2010)
190 established and maintained 19 vegetation plots for wetland mitigation purposes at Juniper Bay.
191 Those 10 m by 10 m plots were located and expanded by 20 m on all sides to create 30 m x 30 m
192 plots for this tree survey, as shown in Figure 1A (plots not drawn to scale). Tree species, height,
193 and diameter at breast height (DBH) were recorded for all trees greater than 10 cm DBH within
194 each vegetation plot. Wood biomass was then estimated for each tree using allometric equations
195 from Gonzalez-Benecke (2011) for loblolly pine (*Pinus taeda* L.) and pond pine (*Pinus serotina*
196 Michx.), and allometric equations from Schroeder et al. (1997) and Jenkins et al. (2003) for all
197 other species. Biomass P content was estimated for all species using P concentrations presented
198 by Bedford et al. (1999). The total plot woody biomass per hectare and woody biomass P per
199 hectare was determined by summing all of the tree biomass and biomass P within each plot, and
200 dividing by the plot area. The total biomass and P_{plant} for the entire Bay was estimated by
201 multiplying the woody biomass per hectare and the woody biomass P per hectare by the area of
202 the Bay.

203 Plant litter (the fibric, Oi soil horizon) samples were collected in October of 2014 from eight
204 randomly selected vegetation plots. A 1 m by 1 m square PVC frame was laid in the center of
205 each plot. Then, five large nails were driven into the ground until the nail head was at the surface
206 of the litter at each of the four corners and one at the center. The litter was then collected for
207 analysis. The heights of each nail above the soil surface were measured and averaged to
208 determine the average depth of plant litter per square meter.

209 The litter was dried in an oven at 70°C for four days, then weighed to determine total litter
210 biomass. Subsamples were ground and analyzed by the Environmental and Agricultural Testing
211 Laboratory at North Carolina State University (Raleigh, North Carolina, USA) for C and P
212 analysis. The percent P by weight (% w/w) for the subsamples was multiplied by the litter dry
213 weight per square meter (g m^{-2}) to estimate the mass of P per square meter (g P m^{-2}) for each site.
214 The P_{lit} for Juniper Bay was estimated by multiplying the average mass of P per square meter by
215 the total area of the Bay (2,910,000 m^2).

216 The perimeter ditch surrounding Juniper Bay drains into a single surface water outflow structure
217 at the edge of the Bay (Figure 1A). Samples from the drainage outlet were taken four times daily
218 from 2010 to 2013 using a Teledyne ISCO automatic water sampler (Teledyne ISCO, Lincoln,
219 Nebraska, USA) and composited into one sample. From 2005 to 2010, manually collected
220 samples (1 L volume) were collected from the center of the channel at the outflow on a monthly
221 basis using a bottle attached to a pole. All water samples were acidified for preservation, and
222 submitted to the North Carolina State University Environmental and Agricultural Testing Service
223 (Raleigh, North Carolina, USA) for DRP and DTP analysis. Only DRP was measured on the
224 grab samples (2005-2010), while both DRP and DTP were analyzed for the daily samples (2010-
225 2013). Organic P (difference between DTP and DRP) was not determined from 2005-2010 and

226 was assumed to contribute to the error term in the P balance as an un-accounted loss. A subset of
227 samples was also analyzed for total P, but no significant difference between total P and DTP was
228 observed. This indicated that particulate P was not present at this site in measureable amounts, so
229 DTP was used for calculating P_{outflow} instead of total P.

230 Surface outflow in the perimeter ditch was measured at the main outlet using two v-notch weirs
231 that were installed in 2001. Discharge rates were measured from December 2010 through 2013.
232 Discharge measurements were made using pressure transducers located upstream and
233 downstream of the weir to determine the stage of the water. Discharge measurements were
234 recorded using a Campbell Scientific CR-10X data logger.

235 Surface water discharge was estimated prior to December 2010 using a monthly water balance.
236 Rainfall was measured on site at three rainfall stations (SI Table 3). Evapotranspiration rates
237 from the MODIS Land Subsets Oak Ridge National Laboratory Distributed Active Archive
238 Center (ORNL DAAC, 2011) were used from January 2005 through December 2013. The
239 MODIS Land Subset ET is remotely sensed ET with a 500 m resolution that is determined from
240 leaf area index (LAI) and radiation at the earth's surface. That dataset provides total ET over 8
241 days. To estimate monthly ET, the 8-day ET values were divided by eight to estimate the daily
242 ET value on the day it was reported. Missing daily ET values were then interpolated using
243 MatLab (MathWorks, Natick, MA, U.S.A.) and summed for each month to estimate total
244 monthly ET from January 2005 to December 2009. Evapotranspiration from January 2010 to
245 November 2010 was estimated using the Thornthwaite method (Thornthwaite 1948) and monthly
246 mean temperature data from the Lumberton Regional Airport (NOAA NCDC 2013)
247 approximately 11 km northwest of Juniper Bay. The total amount of P lost in the drainage water
248 was calculated by multiplying P concentrations in the drainage water by the volume of drainage

249 water leaving through the outflow. This calculation was performed on a daily basis from
250 December 2010 to 2013, and on a monthly basis for 2005 through November 2010.

251 The ΔP_{soil} was determined for an 8-year period, between time zero (date of wetland restoration,
252 2005) and 2013. The error (E) term in Equation 3, based on measured fluxes, was calculated as
253 the remainder term between the soil ΔP_{soil} and flux ΔP .

254 Statistical analysis was performed on the soil total P data using PROC GLIMMIX in SAS 9.3
255 (SAS Institute Inc., Cary, NC, USA) with a gamma distribution. The LSMMeans presented were
256 back-transformed using an “ilink” command. Mehlich-3 extractable P data were analyzed with a
257 natural log transformation using PROC MIXED in SAS 9.3. The LSMMeans reported were back-
258 transformed using a procedure described by Jørgensen and Pedersen (2013). Confidence
259 intervals were corrected for multiple comparisons using a Tukey adjustment. A t-test was
260 performed in SigmaPlot 12.5 (Systat Software, Inc., San Jose, CA, USA) to test for differences
261 in woody biomass and biomass P content between the mineral and organic soils. Summary
262 statistics were determined for P_{atm} using SigmaPlot 12.5.

263 **RESULTS**

264 Total soil P concentrations in 2005 and 2013 are summarized in Table 1. No difference in soil TP
265 was detected between 2005 and 2013 ($p=0.42$) for the entire Bay to a depth of 1m. The organic
266 soils at Juniper Bay had higher ($p<0.0001$) concentrations of TP (0.131 kg m^{-3} , se 0.013) than the
267 mineral soils (0.072 kg m^{-3} , se 0.005) across all three depths and both sampling years. The fixed
268 effect of soil depth was highly significant ($p<0.0001$). The concentrations were highest at the
269 surface and declined with depth. There was no significant difference between years for any soil
270 type-depth combination. Significantly higher TP concentrations were found in the 0-15 cm depth

271 than in the 15-30 cm and 30-100 cm depths for both the mineral and organic soils for both years.
272 There was no significant difference in TP concentration between the 15-30 cm depth and the 30-
273 100 cm depth for either soil in 2005, or in the mineral soil in 2013. However, in 2013 the organic
274 soil had significantly more TP in the 15-30 cm depth than in the 30-100 cm depth. Differences
275 between mineral and organic soil TP concentrations were significant for the 0-15 cm depth and
276 15-30 cm depth, but not the 30-100 cm depth. This was consistent for both sampling years.

277 Rainfall P concentrations during the duration of the study averaged 0.11 mg DTP L⁻¹ (se 0.02).
278 There was no significant difference in P between the three stations. The average concentration of
279 rainfall DTP from 2005 to 2012 was assumed to be equal to the 0.11 mg DTP L⁻¹ observed in
280 this study. That concentration and daily rainfall data for Juniper Bay were used, along with the
281 total area of the Bay, to estimate the P_{atm} following restoration in 2005 – a total of 2.4 Mg (8 kg
282 ha⁻¹) over eight years, or 0.3 Mg (1 kg ha⁻¹) annually. Monthly rainfall is summarized in SI Table
283 3. Estimated monthly P_{atm} is summarized in (Table 2).

284 There was no significant difference in woody biomass or woody biomass P between soil types
285 (mineral versus organic), and the average values across all plots were used for further
286 calculations. The average woody biomass was 29,300 kg ha⁻¹ (se = 5,400), and the average
287 woody biomass P was 26.6 kg P ha⁻¹ (se = 4.9). Extrapolated over the total 291 ha of Juniper
288 Bay, only 7.7 Mg P was extracted by the trees over the eight years following restoration. Woody
289 biomass across Juniper Bay totaled 8,500 Mg. The plant litter biomass and P contents are shown
290 in Table 3. The average P content was 0.32 g P m⁻² (se = 0.07), and the estimated P_{lit} for all of
291 Juniper Bay was 0.93 Mg P (se = 0.21).

292 Discharge rates from Juniper Bay were estimated prior to December 2010 based on a simple
293 water balance as the difference between monthly rainfall (SI Table 3) and monthly ET (SI Table
294 4). Rainfall was compared to normal values for the AgACIS WETS table for the Lumberton
295 Regional Airport which had an observation period of 1971-2000 (USDA-NRCS 2021). Less than
296 normal rainfall was observed for 2005, 2006, 2007, 2011 and 2012, while 2008-2010 had normal
297 rainfall. Remotely measured ET (ORNL DAAC 2011) for 2005-2009, and Thornthwaite-
298 estimated ET rates from 2010-2012 are summarized in SI Table 4. Discharge from the single
299 outflow at Juniper Bay was measured directly starting in December 2010. Monthly discharge
300 rates are summarized in SI Table 5. For months where the estimated discharge was negative
301 (ET>rainfall) the discharge was assumed to be 0 mm.

302 The concentration of DRP over time at the Juniper Bay outflow is shown in Figure 2. The
303 concentration of DRP increased following restoration in 2005, and remained at elevated
304 concentrations until 2010. Following 2010 the concentrations declined to pre-restoration levels.
305 The P discharge, estimated on a monthly basis for 2005-2010 and a daily basis for 2011-2012,
306 totaled 0.5 Mg P. The P balance for Juniper Bay is summarized in Figure 3. The main flux of P
307 entering the Bay during this study was from the atmosphere, at 2.4 Mg P since restoration. Plant
308 uptake into woody biomass was the largest P flux out of the soil, and was estimated at 7.7 Mg P
309 since restoration. Phosphorus that had accumulated in the forest floor litter totaled 0.3 Mg P.
310 Phosphorus leaving the site through the drainage water totaled 0.5 Mg P since restoration. This
311 leaves the error term of -6.1 Mg P, which is larger than all fluxes except Ppl.

312 **DISCUSSION**

313 The primary focus of this study was to determine if Juniper Bay has been, is currently, or will be
314 a source of P for downstream surface waters following restoration of its prior-drained
315 agricultural land. The increase in soil P over 30 years of agricultural fertilization did lead to soil
316 P concentrations significantly higher than un-farmed reference Carolina Bays (Ewing et al. 2012)
317 and a total P pool of 234 Mg P at the time of restoration in 2005. However, no statistical
318 difference was detected between the 234 Mg P in 2008 and 216 Mg P in 2013 the soil P pool.

319 A P balance was estimated in order to better understand P fluxes at Juniper Bay, and to guide
320 future management. The main flux of P into the Bay was P_{atm} . Atmospheric deposition of P was
321 small, at 2 Mg P over 8 years. The concentration of DTP in the rainwater was also approximately
322 the same concentration as in the drainage water. Because the runoff ratio (ratio of runoff to total
323 rainfall) of Juniper Bay is very small, very little of that rain (and P) actually reached the outflow
324 structure. The flux of P out of Juniper Bay in the drainage water was under 0.5 Mg P since
325 restoration. The largest P loss was due to plant uptake. The trees at Juniper Bay acquired 7.7 Mg
326 of P since restoration, and resulted in an additional accumulation of 0.3 Mg P into plant litter.
327 This flux of P from the soil and into woody biomass and plant litter should slow any potential
328 release of P to drainage waters.

329 The error term of 6.1 Mg P of loss is larger than all of the other fluxes except plant uptake. One
330 potential source of error was having only DRP and not DTP measurements of the drainage water
331 prior to 2010. The organic P missed may account for a larger flux of P out of the wetland.
332 However, the measured flux of P in the drainage water was still very small, and even doubling
333 that flux would only increase it to 1 Mg P over 8 years. Plant uptake is likely larger than was

334 predicted due to P taken up by trees <10 cm DBH, shrubs, and herbaceous plants, which were
335 not measured in this study.

336 The agreement of measured runoff volumes at the outflow with runoff predicted with a water
337 balance suggest that losses or gains of water from the surrounding landscape are relatively
338 minimal (see supplemental information). This is further evidenced by the small hydrologic
339 gradients in four transects around the Bay (SI Table 1), and the resulting relatively-slow
340 porewater velocities (SI Table 2). However, further study of the hydrology of Juniper Bay and
341 the surrounding landscape, and of P concentrations in underlying aquifers may be warranted if a
342 reduction of the P balance error term is desired.

343 While post-restoration concentrations of P in the Juniper Bay drainage water did depict a small
344 release of P out of Juniper Bay, concentrations have since declined to pre-restoration levels (\leq
345 0.1 mg P L^{-1}) within five years of restoration (Figure 2). Phosphorus concentrations above 0.1
346 mg L^{-1} would be expected to contribute to eutrophication in freshwater systems (Correll 1998).
347 The concentration of P in the drainage water was also equal to P concentrations found in the
348 rainwater. In addition, the 0.5 Mg P that has left Juniper Bay through the outflow since
349 restoration accounts for approximately 0.2% of the total amount of P at the site in 2005. Because
350 of the low concentrations of P in the drainage water, and the low magnitude of P losses to the
351 drainage water relative to the total P pool, P export from Juniper Bay to surface waters is not
352 expected to be a major concern in the future. Bruland et al. (2003) determined that in a restored
353 Carolina Bay wetland complex the export of soluble reactive phosphorus and total phosphorus
354 from the restored wetland was less than that of an actively farmed wetland, thus concluding that
355 wetland restoration after just two years resulted in a net improvement to water quality. While this
356 study did not include measurements of phosphorus export during active agricultural production

357 in Juniper Bay, concentrations of phosphorus following restoration did eventually decrease to
358 post-agricultural production prior to restoration of wetland hydrology. Ewing et al. (2012)
359 estimated that Juniper Bay had three times as much extractable P than in nearby reference
360 Carolina Bays. If that proportion of extractable P is also true for total P, and if Juniper Bay
361 continues to lose total P at the rate of approximately 1% per year, then it will take at least 60
362 years for Juniper Bay to return to “natural” P concentrations. Plant uptake may reduce the
363 amount of plant available P – the P fraction most easily exported. However P loss to plant uptake
364 alone will take decades to centuries to reduce soil P concentrations down to natural
365 concentrations.

366 The results of this study indicate that while most of the residual soil P that was left over from
367 agricultural production is still in the Juniper Bay soils, it is not moving off site. This indicates
368 that Carolina Bays, like Juniper Bay, make excellent potential sites for wetland restoration.
369 However, Carolina Bays that are drained by streams may be exceptions to this rule. Such
370 wetlands would be expected to have more P leaving the site through surface outflow because of
371 higher hydraulic gradients caused by the dissecting streams. The ideal areas for wetland
372 restoration are the closed depressions that have precipitation as the main water source and
373 evapotranspiration as the main water loss.

374 In weighing the potential risks and benefits of restoring Carolina Bay wetlands that have been
375 previously been used for production agriculture, it is important to consider the conversions
376 between land uses for these wetlands within the region. In a concurrent study, separate from that
377 presented here, Sullivan et al. (2017) inventoried Carolina Bays in Bladen, County, North
378 Carolina a representative “Bay-dense” region within the southeastern Coastal Plain close to
379 Robeson, County, North Carolina where the present study was conducted . They documented

380 land-use change from 1972 through 2010 using decadal Landsat imagery. They found that during
381 that time period, 43% of Bays and 91% of Bay area was associated with land-use change
382 between 1972 and 2010. In 1972, Bays were predominantly forested (79% by count); with
383 remaining Bays converted to agriculture or urban use prior to 1972. Land-use changes were
384 predominantly from forest to agriculture (46%) and agriculture to forest (37%). Conversion to
385 forest remained low from 1984 to 1991 and from 2000 to 2010, with a net loss in agricultural
386 land use of 2,085 and 1,457 ha, respectively. A surge in conversion from forest to agriculture
387 occurred between 1991 and 2000, which was surprising given the 1990 US Army Corps of
388 Engineers/Environmental Protection Agency agreement targeting no net wetland loss. From
389 1972 through 2010, there was an estimated net gain of 744 ha of Bay forest relative to Bay
390 agriculture. Sullivan et al. (2019) conducted a follow-up study assessing the risk of phosphorus
391 export to nearby or intersecting streams. They found that 1360 Carolina Bays in Bladen County,
392 North Carolina representing 43% of the Bays and 80% of the total Bay area had streams that
393 either intersected the Bays or came within 15 m of the edge of a the Bays. These wetlands posed
394 a risk of P export depending on the land use or changes in land use of each Bay. Isolated
395 wetlands without a nearby or intersecting stream were determined to pose little risk of P export.

396 **SUMMARY & CONCLUSIONS**

397 The objective of this study was to create a P balance for a Carolina Bay restored from
398 agricultural land. Juniper Bay was restored by filling in primary ditches, plugging secondary
399 ditches, and maintaining the perimeter ditch surrounding the site. This resulted in a low hydraulic
400 gradient for drainage water. The soil P pools were determined to be 234 Mg in 2005 and 216 Mg
401 in 2013, though no significant difference was detected. Phosphorus fluxes into and out of Juniper
402 Bay included a gain in P from atmospheric deposition, and losses of P to surface water outflow

403 and plant uptake. The error term depicts an unaccounted loss of 6.1 Mg P, which was larger than
404 all fluxes except plant uptake by trees. Phosphorus loss to surface waters was minimal both in
405 magnitude (0.5 Mg P over eight years), and in current concentrations (approximately 0.1 mg
406 P/L). That concentration of P exiting the Bay is approximately the same as was found in
407 rainwater at Juniper Bay, and is not expected to contribute to eutrophication of downstream
408 surface waters. Contributions of water and P to and from the surrounding landscape were
409 determined to be minimal based on a comparison of observed runoff values with those predicted
410 from a water balance, and from an evaluation of existing transects of P concentration and
411 hydrologic gradients around the perimeter of Juniper Bay. A P balance for Juniper Bay was
412 estimated based on measured changes in total P and fluxes of P. It will likely take 60 years or
413 longer for Juniper Bay to return to “natural” concentrations of P. There is a possibility that future
414 restorations of Carolina Bays from drained agricultural production back to wetlands may
415 contribute agrichemicals to nearby streams and drainageways. However, in regard to P, if
416 restoration methods as described for Juniper Bay are employed, impairment of nearby streams
417 and drainageways is unlikely.

418 Based on these results, we conclude that Carolina Bay wetlands similar to Juniper Bay are
419 promising sites for wetland restorations due to the low risk of P contributions following
420 restoration from agricultural production. The broad, nearly level Carolina Bays result in very low
421 hydraulic gradients following restoration back to wetlands, thus reducing movement of P to
422 nearby streams or drainageways. However, internal drainage ditches must be filled and/or
423 plugged during the restoration process. Carolina Bays that are deeply dissected by streams or
424 ditches may have sufficient hydraulic gradients to facility P transport, so restoration of such sites
425 must be done with care.

426 **DECLARATIONS**

427 **Funding**

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429 Carolina System, and by the US Department of Agriculture - Agriculture and Food Research
430 Initiative.

431 **Conflicts of Interest**

432 The authors have no conflicts of interest.

433 **Availability of Data and Material**

434 The datasets generated during and/or analyzed during the current study are available from the
435 corresponding author on reasonable request.

436 **Code Availability**

437 The code used during the current study is available via Moorberg (2014)

438 **Authors' Contributions**

439 CJM lead efforts for sample collection, sample analysis, data analysis, data presentation, and
440 writing. MJV was the principle investigator for both sources of funding, supervised the research
441 activities of CJM, and was a major contributor to writing the manuscript. CPN contributed to
442 sample collection and analysis. JGW assisted with sample collection for the archived soil
443 samples. DDR assisted soil total P analysis. All authors provided revisions to manuscript drafts
444 and read and approved the final draft.

445 **Ethics Approval**

446 Not applicable

447 **Consent to Participate**

448 Not applicable

449 **Consent for Publication**

450 Not applicable

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- 566

567 **TABLES**

568 Table 1. Soil TP concentrations by soil type, depth, and year.

Soil	Depth	2005			2013		
		LSMean	se	Significance	LSMean	se	Significance
	cm	kg TP m ⁻³		a b c	kg TP m ⁻³		a b c
Mineral	15	0.122	± 0.013	A A A	0.132	± 0.015	A A A
Mineral	30	0.069	± 0.008	A B A	0.058	± 0.007	A B A
Mineral	100	0.051	± 0.007	A B A	0.044	± 0.006	A B A
Organic	15	0.257	± 0.039	A A B	0.207	± 0.032	A A B
Organic	30	0.118	± 0.018	A B B	0.160	± 0.025	A B B
Organic	100	0.076	± 0.017	A B A	0.065	± 0.015	A C A

569 ^aComparison of TP concentration between years for a given soil type and depth in column

570 ^bComparison of TP concentration between depths within a given year and soil type in column

571 ^cComparison of TP concentration between soil types within a given year at a given depth in
572 column

573 Means with the same letter not significantly different ($\alpha=0.05$).

574

575 Table 2. Monthly P_{atm} deposition (kg TP) between January 2005 and December 2012.

Month	2005 ^a	2006	2007	2008	2009	2010	2011	2012
Jan	16.3	22.3	5.2	23.0	10.3	26.3	10.9	22.4
Feb	13.4	22.6	14.1	33.8	13.6	33.7	38.6	27.7
Mar	15.7	4.5	11.5	30.2	27.1	26.0	32.6	29.4
Apr	16.1	13.6	20.5	29.4	8.8	5.5	19.6	17.1
May	11.9	40.0	13.1	23.7	76.1	24.7	29.8	49.9
Jun	15.3	42.6	26.9	22.6	36.9	54.2	12.0	33.3
Jul	36.7	35.6	4.3	28.0	32.4	80.5	28.7	28.8
Aug	4.4	44.8	19.3	52.1	47.1	23.9	57.6	68.8
Sep	10.9	8.3	3.5	66.7	2.6	61.7	29.5	26.3
Oct	19.5	5.6	14.0	6.5	21.8	9.8	17.7	58.0
Nov	26.2	28.3	0.7	37.3	56.5	10.3	26.3	0.1
Dec	15.8	23.1	32.8	23.2	48.0	18.5	5.8	22.3
Total	202.1	291.2	165.8	376.6	381.2	375.2	309.0	581 329.8

582 ^aRainfall data was acquired from a nearby weather station at the Lumberton, NC airport (NOAA
 583 NCDC 2013).

584

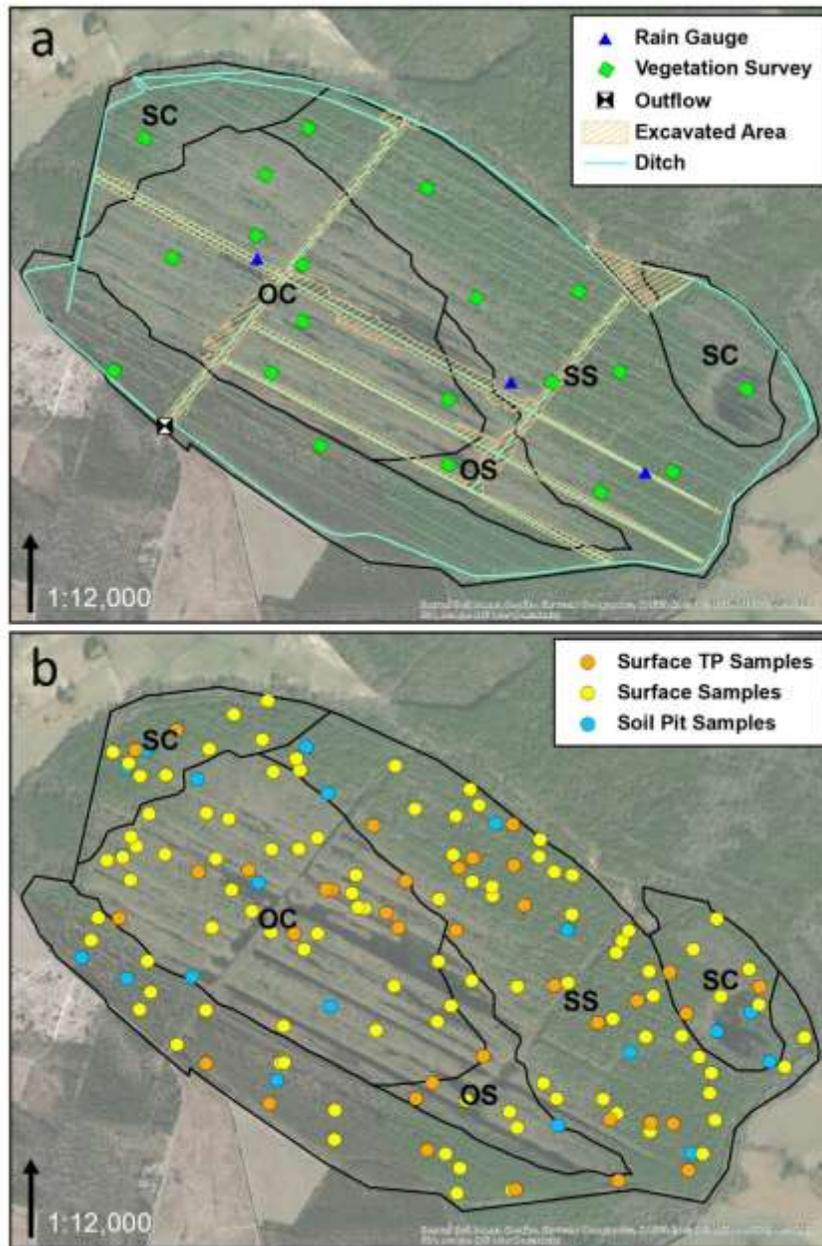
585 Table 3. Plant litter biomass and P content.

Plot	Dominant Trees Present	Average Litter Depth (cm)	Litter Dry Weight	P Concentration	P Content
		cm	g	% (w/w)	g P/m
1	Pine	4.50	850.6	0.08	0.71
8	Bald Cypress	2.60	337.7	0.09	0.30
9	Pine, Oak, Sweetgum	4.82	870.4	0.05	0.46
12	Bald Cypress, Pine, Willow	2.76	274.4	0.05	0.14
14	Pine, Bay	1.38	259.5	0.07	0.18
15	Bay, Bald Cypress	2.58	290.5	0.10	0.30
16	Oak	3.90	488.8	0.07	0.36
19	Bald Cypress	0.76	82.5	0.11	0.09
Average P Content					0.32 (se 0.07)

586

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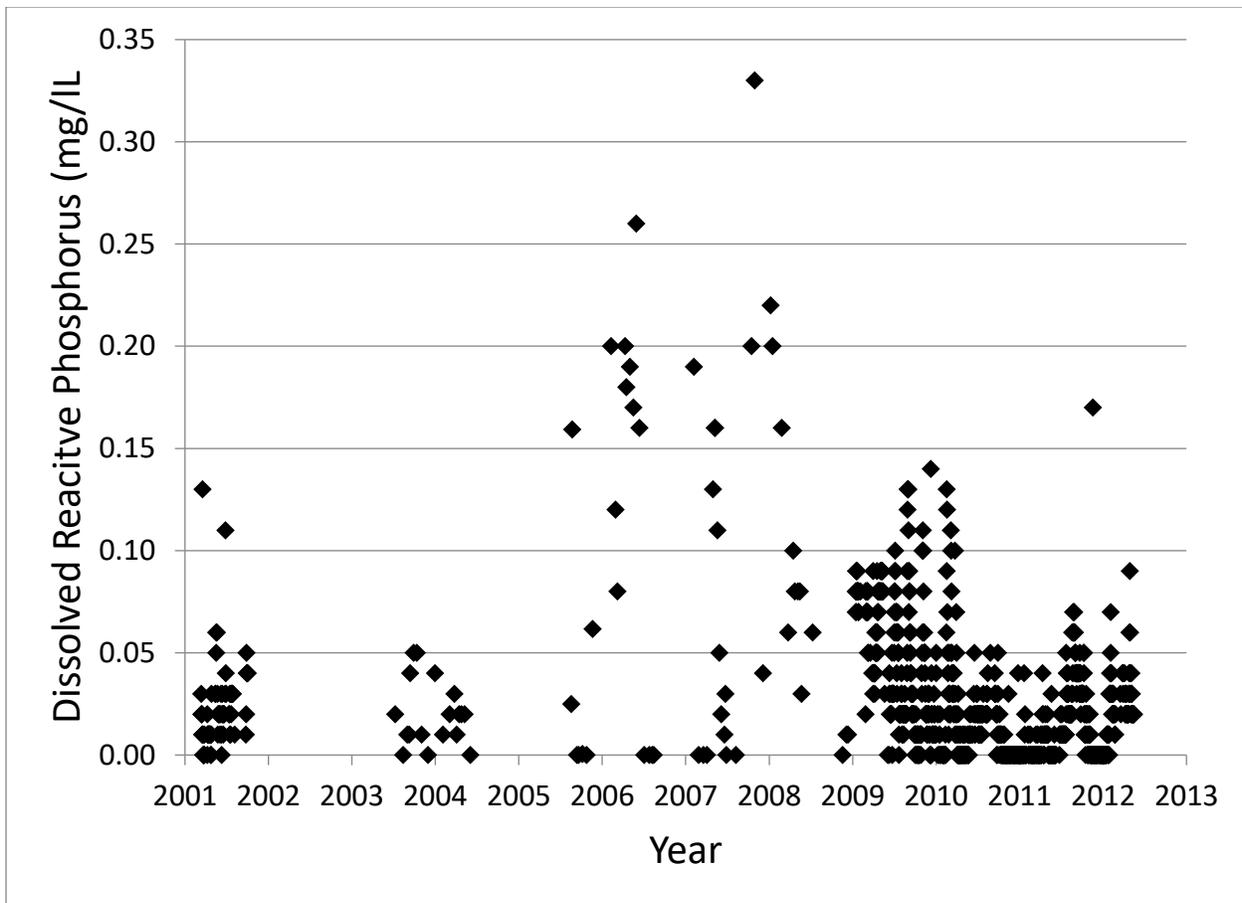
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590

591 Figure 1. Maps of Juniper Bay depicting locations of a) previous and existing drainage
 592 ditches, perimeter ditch outflow, vegetation survey locations (not drawn to scale), rain
 593 gauge location, and mineral and organic soil distribution; and b) resampled surface soil
 594 locations for extractable and total P, and resampled soil pit locations. The mapping units
 595 depict four soil conditions, including sands over clayey subsoil (SC), sands over sandy
 596 subsoil (SS), organic soil over clayey subsoil (OC), and organic soil over sandy subsoil
 597 (OS).

598

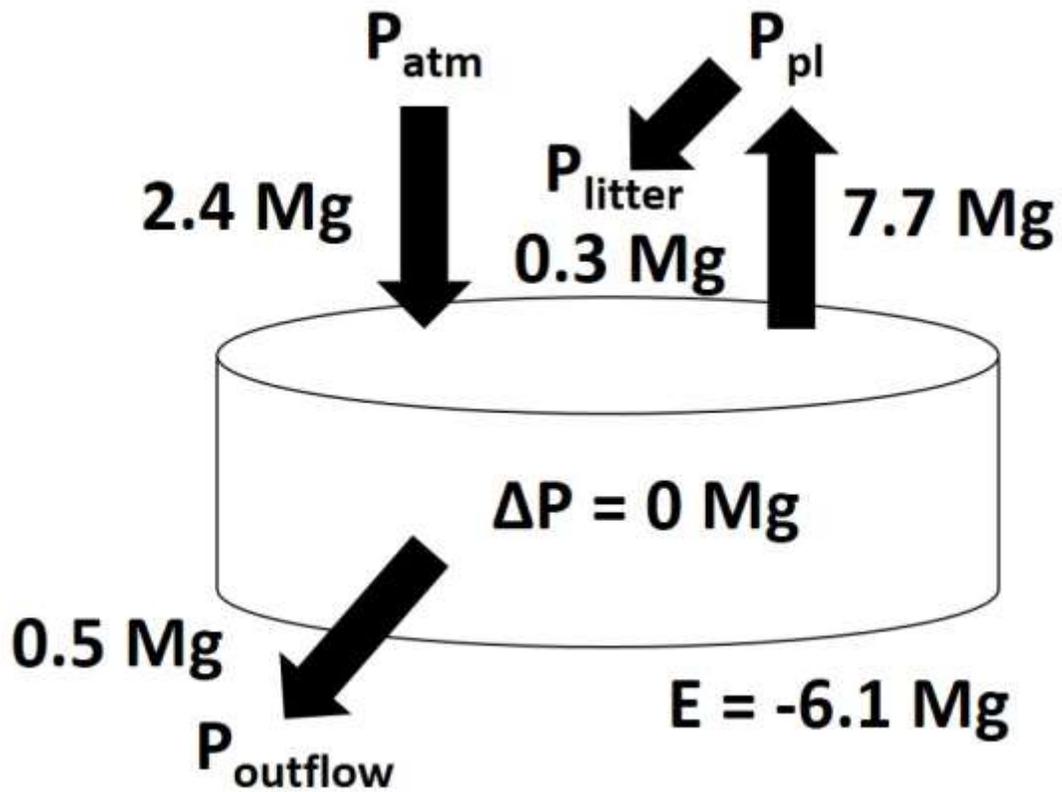


599

600 Figure 2. Concentration of DRP at the Juniper Bay outflow over time. The wetland was
 601 restored in 2005, after which an increase in DRP at the outflow was observed through
 602 approximately 2010. The DRP concentrations declined to pre-restoration levels
 603 thereafter. Eutrophication would be expected at 0.10 mg DRP per liter in freshwaters,
 604 assuming N is not limiting. Outflow concentrations only exceeded that concentration
 605 once after 2010.

606

607



608

609 Figure 3. Summary of the P balance for Juniper Bay from 2005-2013. The fluxes include
 610 change in total P (ΔP_{soil}), atmospheric deposition (P_{atm}), plant uptake (P_{pl}), plant
 611 litter (P_{litter}), surface water outflow ($P_{outflow}$), and error (E).

612

613 **LIST OF SUPPLEMENTAL TABLES**

614 SI Table 1. Water Balance Test of Hydrology Assumptions.

615 SI Table 2. Estimations of Flow for Each Transect and Sand Layer. Porewater velocity
616 was estimated from Hydraulic gradients and soil properties for the four piezometer
617 transects at Juniper Bay. The saturated conductivities used were reported by Pati
618 (2006). Porosity was estimated from average bulk densities reported by Ewing (2003).
619 Hydraulic gradients were calculated from measurements made on May 15, 2013. Flow
620 rates were small overall, due to both the small gradients present, and the low
621 conductivities.

622 SI Table 3. Monthly Rainfall from January 2005 to December 2012.

623 SI Table 4. Monthly ET between January 2005 and December 2012. Evapotranspiration
624 was measured remotely for 2005-2009, and estimated using the Thornthwaite equation
625 in from 2010 to 2012.

626 SI Table 5. Monthly runoff between January 2005 and December 2012. Runoff was
627 estimated using a simple water balance for 2005-2010, and measured directly for 2011
628 and 2012. For estimations of P, months with negative estimated runoff were assumed to
629 have no runoff.

630 **LIST OF SUPPLEMENTAL FIGURES**

631 SI Figure 1. Piezometer Transect Across the Southwest Rim of Juniper Bay.

632 SI Figure 2. Piezometer Transect Across the Southeast Rim of Juniper Bay.

633 SI Figure 3. Piezometer Transect Across the Northeast Rim of Juniper Bay.

634 SI Figure 4. Piezometer Transect Across the Northwest Rim of Juniper Bay.

635 SI Figure 5. The apparatus used for collection of atmospheric deposition of P, alongside
636 a tipping bucket rain gauge and a traditional rain gauge. The sample collection bottle is
637 not shown.

Figures

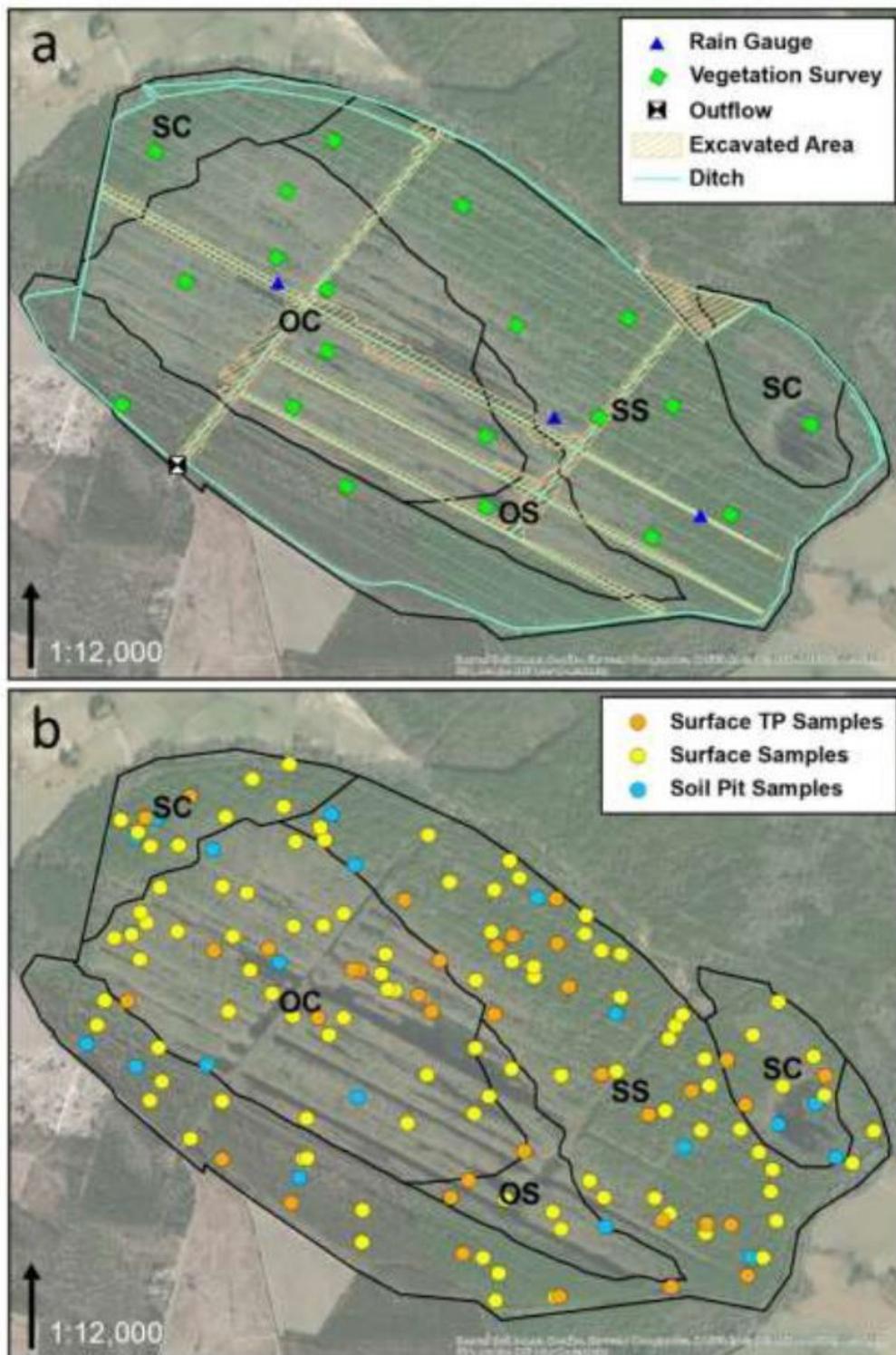


Figure 1

Maps of Juniper Bay depicting locations of a) previous and existing drainage ditches, perimeter ditch outflow, vegetation survey locations (not drawn to scale), rain gauge location, and mineral and organic soil distribution; and b) resampled surface soil locations for extractable and total P, and resampled soil pit

locations. The mapping units depict four soil conditions, including sands over clayey subsoil (SC), sands over sandy subsoil (SS), organic soil over clayey subsoil (OC), and organic soil over sandy subsoil (OS). Note: The designations employed and the presentation of the material on this map do not imply the expression of any opinion whatsoever on the part of Research Square concerning the legal status of any country, territory, city or area or of its authorities, or concerning the delimitation of its frontiers or boundaries. This map has been provided by the authors.

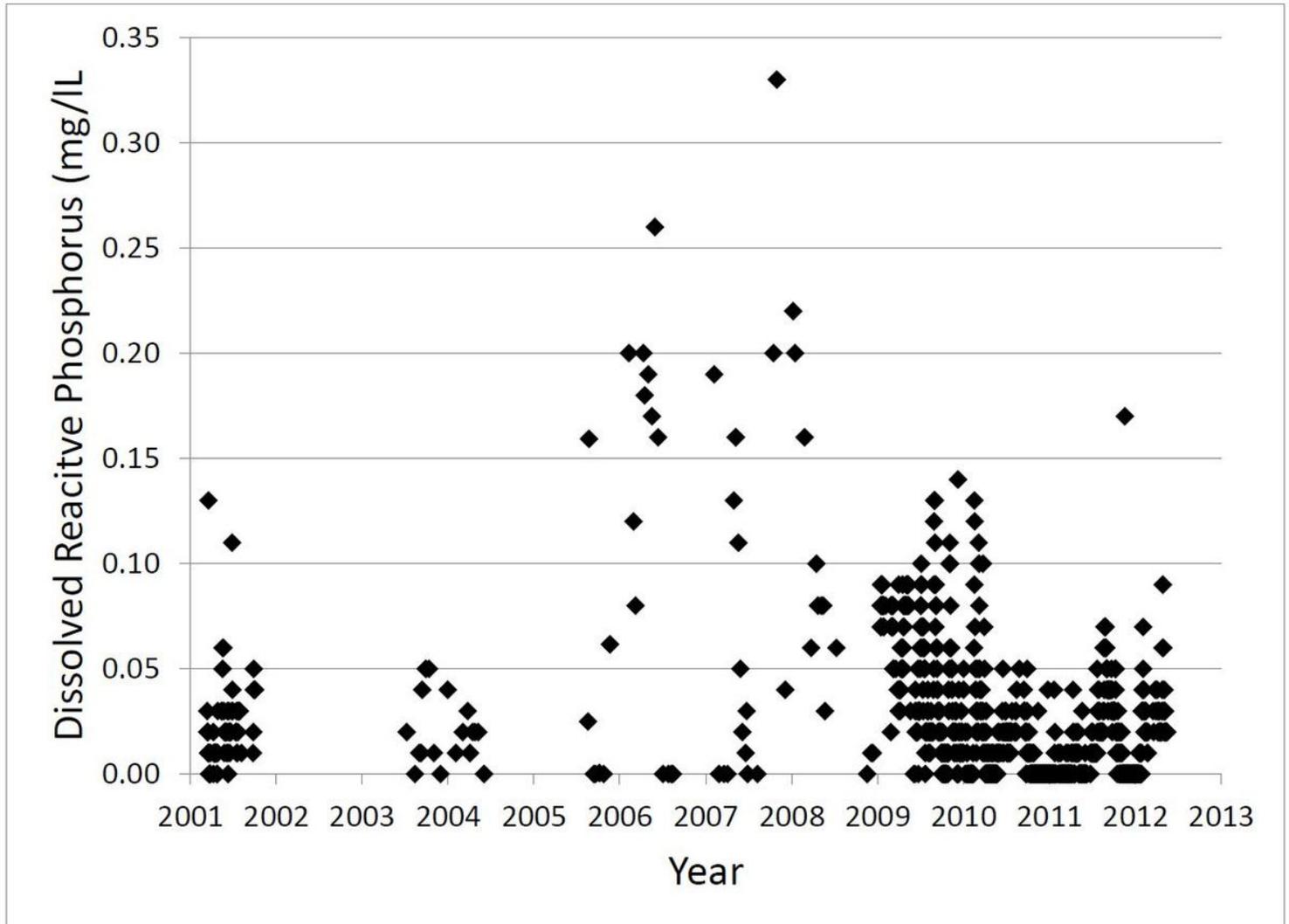


Figure 2

Concentration of DRP at the Juniper Bay outflow over time. The wetland was restored in 2005, after which an increase in DRP at the outflow was observed through approximately 2010. The DRP concentrations declined to pre-restoration levels thereafter. Eutrophication would be expected at 0.10 mg DRP per liter in freshwaters, assuming N is not limiting. Outflow concentrations only exceeded that concentration once after 2010.

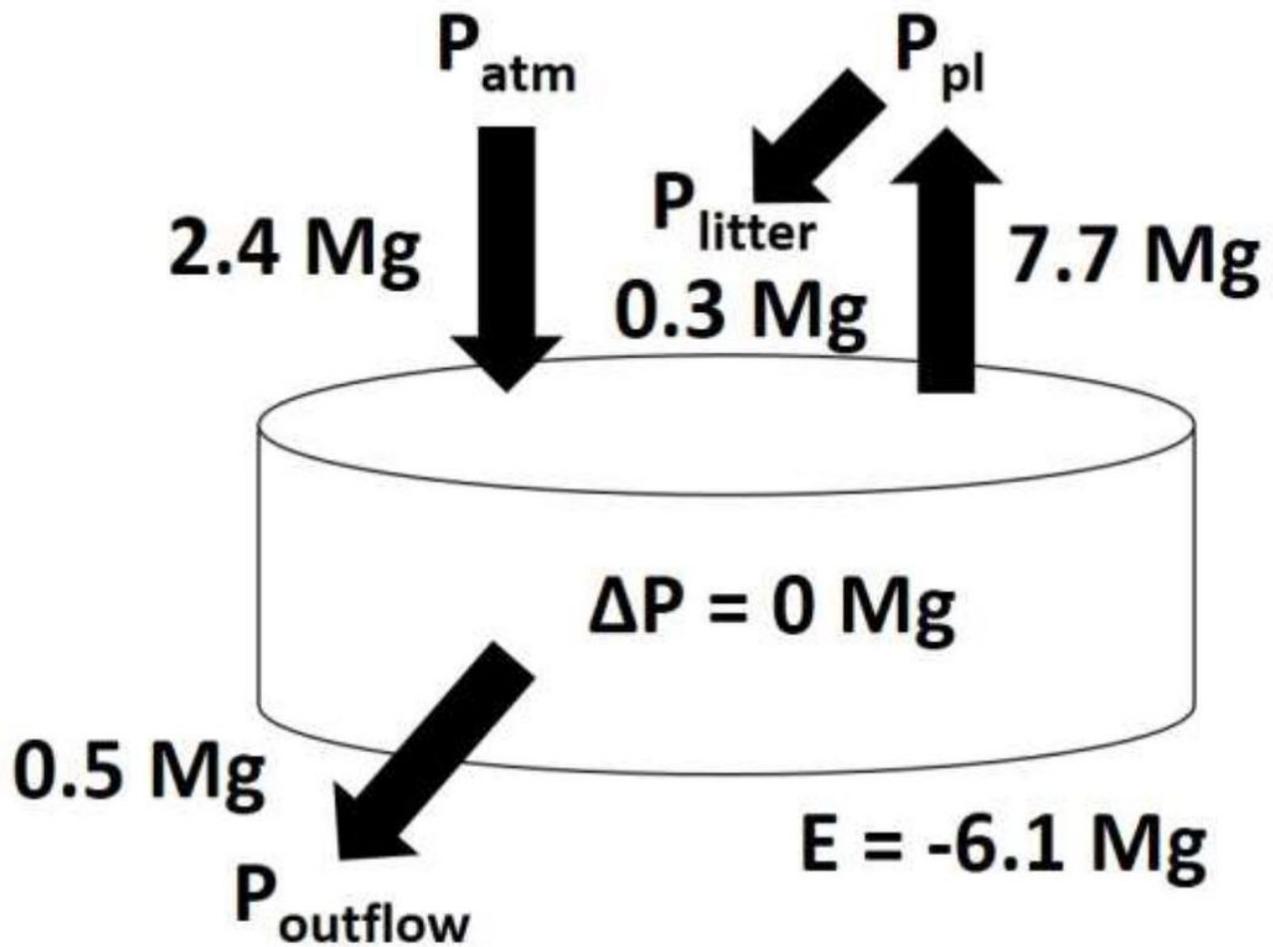


Figure 3

Summary of the P balance for Juniper Bay from 2005-2013. The fluxes include change in total P (ΔP_{soil}), atmospheric deposition (P_{atm}), plant uptake (P_{pl}), plant litter (P_{litter}), surface water outflow ($P_{outflow}$), and error (E).

Supplementary Files

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- [PhosphorusBalanceWetlandsSubmissionSI.pdf](#)