

Reintroduction of salt marsh vegetation and phosphorus fertilisation improve plant colonisation on seawater-contaminated cutover bogs

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SUMMARY

Coastal bogs that are used for peat extraction are prone to contamination by seawater during storm events. Once contaminated, they remain mostly bare because of the combination of high salinity, low pH, high water table and low nutrient availability. The goal of this research was to investigate how plant colonisation at salt-contaminated bogs can be accelerated, in order to prevent erosion and fluvial export of the peat. At two seawater-contaminated bogs, we tested the application of rock phosphate and dolomitic lime in combination with five plant introduction treatments: transplantation of *Carex paleacea*; transplantation of *Spartina pectinata*; transfer of salt marsh diaspores in July; transfer of salt marsh diaspores in August; and no treatment (control). The effects of different doses of lime on the growth of *C. paleacea* and *S. pectinata* were also investigated in a greenhouse experiment. In the field, phosphorus fertilisation improved plant growth. Transplantation of *C. paleacea* resulted in the highest plant colonisation, whereas salt marsh diaspore transfer led to the highest species diversity. Lime applications did not improve plant establishment in either the field or the greenhouse. To promote revegetation of seawater-contaminated cutover bogs, adding P is an asset, *Carex paleacea* is a good species to transplant, and the transfer of salt marsh diaspores improves plant diversity.

KEY WORDS: rehabilitation, revegetation, sedge transplantation, seed bank, vacuum milled peatlands

INTRODUCTION

Many coastal bogs in eastern Canada are used for vacuum milling (extraction) of horticultural peat, which lowers their surfaces and renders them prone to invasion by seawater. In the future, this tendency could be exacerbated by a rise in sea level resulting from global warming (Warrick & Oerlemans 1990). Bogs that are subject to ongoing contamination by seawater after horticultural peat extraction often remain mostly bare of vegetation for several decades (Figure A1, Appendix). It is impossible to restore these sites back to *Sphagnum* dominated ecosystems because *Sphagnum* mosses cannot successfully perform vital exchanges with their environment when salt is present, even at low concentrations (Clymo 1963, Wilcox 1984). Impediments to the establishment and growth of other plants include not only salinity (Bernstein & Hayward 1958, Greenway 1962, Greenway & Munns 1980, Wyn Jones & Gorham 2004, Breathnach 2008), but also low pH, high water table (Grable 1966, Barrett-Lennard 2003, Gibbs & Greenway 2003) and low nutrient availability. The identification of species that can tolerate these harsh conditions is an essential step towards achieving the revegetation of cutover bogs that have been contaminated by seawater.

Upper salt marsh is a natural ecosystem with many similarities to seawater-contaminated cutover peatland, in that both are saline wetlands which are not inundated by daily flood tides. Therefore, it seems possible that salt marsh plants could be used to revegetate cutover bogs that have suffered seawater contamination. Although the salinity of seawater-contaminated bogs is, on average, lower than that of salt marshes, the capacity of salt marsh species to colonise these sites should not be affected, since most salt marsh plants can also grow in freshwater habitats (Barbour 1970, Waisel 1972). *Spartina pectinata* Link. and *Carex paleacea* Schreb. ex Wahlenb. are found in abundance in upper salt marsh systems. The genus *Spartina* is often used in salt marsh restoration because it is a strong coloniser (Broome *et al.* 1988). *C. paleacea*, a member of the *Cyperaceae* family, is often found in bogs (Broome *et al.* 1975, Bruederle & Fairbrothers 1986), making it a good candidate for testing. Preliminary trials in cutover bogs that had been contaminated by seawater showed that both species were able to establish in the harsh waterlogged and salty conditions, but did not proliferate outside the area of reintroduction (Breathnach 2008, Montemayor *et al.* 2008, Montemayor *et al.* 2015). According to these studies, *S. pectinata* and *C. paleacea* are good choices for

continued testing on seawater-contaminated bogs but their propagation success needs to be improved, along with plant diversity.

Apart from transplanting vascular plants, which requires much handling and is time consuming, another option to increase plant diversity is to adapt the moss layer transfer technique which has been used to restore extracted bogs across northern America (Quinty & Rochefort 2003). This method involves collecting the uppermost 10 cm of moss carpet (which also include seeds, roots and rhizomes of vascular plants) from a natural bog (donor site) and spreading it on the surface of the extracted bog. Various typical bog plant species are reintroduced along with *Sphagnum* mosses (Poulin *et al.* 2013). In the case of seawater-contaminated bogs, a salt marsh could be used as the donor site.

Mineral fertilisers are often used to ensure successful propagation of introduced plant species. Salt marsh restoration procedures often include the addition of nitrogen (N) (Squiers & Good 1974, Cargill & Jefferies 1984, Zedler 1984, Broome *et al.* 1988, Weishar *et al.* 2005, Konisky *et al.* 2006). Mineral fertilisers could be used to mitigate the differences in nutrient availability between bogs and salt marshes. Phosphorus (P) (Sottocornola *et al.* 2007, Breathnach 2008, Andersen *et al.* 2011), calcium (Ca) and magnesium (Mg) (Raven *et al.* 2003, Andersen *et al.* 2011) have been identified as the main limiting nutrients in organic soils, and the availability of nutrients in bogs is generally lower than in salt marshes (Valiela *et al.* 1978, Craft *et al.* 1999, Deegan *et al.* 2007, Andersen *et al.* 2011) even after seawater intrusion (Montemayor *et al.* 2010). Previous trials carried out in seawater-contaminated bog remnants showed that rock phosphate added at a rate of 25 g m⁻² did not significantly increase the cover of introduced plants, but an accidental spill of fertiliser in a pond resulted in a good revegetation of the edges of the pond suggesting that a higher dose of rock phosphate might be required to improve plant growth (Breathnach 2008). Increasing soil pH by adding lime could also improve the availability of P and other nutrients, provide the two limiting nutrients Ca and Mg, and lower the availability of potentially toxic micro-elements such as Al at the same time (Brady & Weil 2003).

The objective of this study was to develop a revegetation approach for coastal cutover bogs contaminated with seawater by testing the introduction of different plant materials in combination with fertiliser applications. As a secondary objective, we tested the effect of lime addition on the germination, seedling survival and growth of *S. pectinata* and *C. paleacea*.

METHODS

Large scale field experiment

Study sites

The field sites were two coastal bogs located near the townships of Pokesudie and Shippagan in the province of New Brunswick, Canada (47° 49' N, 64° 49' W and 47° 41' N, 64° 46' W, respectively). Both sites present surface altitudes between 0 and 2 metres above sea level and were invaded by seawater during a storm surge in January 2000, after which peat extraction ceased (Figure A2). Berms and ditches were built around both bogs to prevent further invasion by seawater. The berms at the Shippagan site failed, with the result that it continued to receive seawater periodically, during storm events and high tides. Following seawater contamination and the abandonment of peat extraction activities, drainage ditches collapsed naturally over time. By the time the present study began, the water table remained near or at the peat surface during most of the growing season (May to September).

Experimental design

To test the establishment of local marsh plants in seawater-contaminated cutover bogs, a field trial was designed as a factorial experiment comprising four blocks and three factors: 1) plant material introduced (5 types), 2) liming treatment (+/-) and 3) phosphorus fertilisation (+/-) treatment. The 9 m² plots installed in 2011 were at least five metres from each other to prevent lateral movement of fertilisers due to high water table level (7 cm below the surface on average). The field experiment was set up in those parts of the bogs that had been most severely affected by seawater (surrounded by dotted lines in Figure A2) and lasted two growing seasons. The growing season of 2011 was wet (621 ± 19 mm) while 2012 (461 ± 11 mm) was closer to historical standards (412 ± 18 mm) according to data from the meteorological stations at Caraquet (47° 48' 00" N, 64° 52' 00" W) and Bas Caraquet (47° 48' 08" N, 64° 50' 00" W) (Environment Canada 2013).

Plant material

The plant treatments were: 1) reintroduction of *S. pectinata*, 2) reintroduction of *C. paleacea*, 3) diaspore transfer of salt marsh layer in mid-July, 4) diaspore transfer of salt marsh layer in mid-August and 5) a bare peat control. The donor site was the natural salt marsh located next to the bog in each case. The main soil characteristics of the salt marshes and bogs are presented in Table 1. For individual species transplantations, 49 uniform 5 cm diameter

Table 1. Soil environmental conditions in sections of the seawater-contaminated Pokesudie and Shippagan peatlands and in the nearby salt marshes where plant material was collected (donor sites). P, Ca and Mg concentrations are included to show how the soils differed with respect to these nutrients before fertiliser additions aiming to increase nutrient availability. The values are means with (SE) in parentheses.

Environmental conditions	Northern Pokesudie bog	North-eastern Shippagan bog	Pokesudie salt marsh (donor site)	Shippagan salt marsh (donor site)
Salinity (‰)	0.48 (0.03)	1.39 (0.07)	7.03 (1.06)	7.63 (0.87)
pH	3.6 (0.0)	3.5 (0.0)	5.7 (3.0)	5.2 (0.1)
Redox (mV)	173 (6)	143 (8)	41 (32)	55 (36)
Water table level (cm)	-7 (1)	-5 (1)	-9 (1)	-11 (3)
[P] ($\mu\text{g g}^{-1}$)	11.0 (0.2)	0.6 (0.2)	81.0 (0.9)	21.0 (0.3)
[Ca] (mg g^{-1})	1.4 (0.1)	1.5 (0.1)	2.2 (0.8)	0.7 (0.3)
[Mg] (mg g^{-1})	1.8 (0.1)	3.0 (0.2)	5.3 (1.5)	1.3 (0.2)

plugs (rhizome and soil) of *C. paleacea* and *S. pectinata* were collected from the salt marsh and planted in each plot at the end of June 2011. The salt marsh diaspore layer was transferred in mid-July and mid-August 2011, by first detaching and cutting plant material into small pieces using a rototiller. This material, which included aerial and underground tissues along with seed bank, was collected by hand and then spread on the contaminated cutover bog (Figure 1). Plant material collected from 1 m² of salt marsh was spread over 6 m² of experimental plots.

Liming and phosphorus fertilisation treatments

In the transplant plots of *C. paleacea* and *S. pectinata* and the control plots (with no plant material), dolomitic lime ($\text{CaCO}_3\cdot\text{MgCO}_3$; 18 or 0 g) or rock phosphate (H_2PO_4 ; 9 or 0 g) was placed in the planting holes. In the marsh diaspore transfer technique, lime (100 or 0 g m⁻²) or rock phosphate (50 or 0 g m⁻²) was spread on the surfaces of the plots and incorporated into the peat with a rake just before spreading the diaspore material.

Monitoring

All monitored variables were measured in a 2 × 2 m quadrat centred within each 9 m² experimental plot. Abiotic (pH, salinity, redox, water table level, peat chemistry) and biotic (height, percent cover of introduced and spontaneously colonising species, aerial and underground biomass, leaf tissue analysis) data were recorded in mid-July 2012 in all experimental plots. *Spartina pectinata* (very

abundant) and *Spartina alterniflora* Loisel. (scarce) were recorded as *Spartina sp.* because inflorescences were often absent at the time of survey which makes species identification difficult. Furthermore, hybridisation is very frequent in the genus *Spartina*, and this makes species identification more hazardous (Ainouche *et al.* 2004). Aerial and underground biomass was collected from the centre of each plot in August 2012. Quadrats of 50 × 50 cm were used for aerial biomass and 25 × 25 cm for underground biomass. Aerial biomass was sorted to separate spontaneous vegetation and introduced plants before drying and weighing. For chemical analyses of leaves, only material from transplants of *C. paleacea* and *S. pectinata* was analysed, since material from the two diaspore transfer treatments was too diverse (see Table A1 in Appendix for a description of the composition of plant cover in these plots). Digestion of foliar and peat tissues was carried out according to the wet oxidation procedure of Parkinson & Allen (1975). Exchangeable nutrients were extracted using 0.1 M $\text{NH}_4\text{Cl}\text{-BaCl}_2$ (Amacher *et al.* 1990), P was extracted using P Bray II (Bray & Kurtz 1945) with Quickem method 12-115-01-1A, and total N was extracted using Quickem method 13-107-06-2D. A Quickem 4000 (Lachat Instruments, Milwaukee, WI, USA) was used for the analyses.

Data analyses

Variables were analysed by three-way ANOVA using the MIXED procedure of SAS (Statistical Analysis System 9.3, SAS Institute Inc., Cary, NC,



Figure 1. Collection of diaspore material from the salt marsh at Pokesudie. The material was collected by hand after a rototiller had been passed over the donor area (left). The picture on the right is a close-up of the diaspore material, which includes the aerial parts of the vegetation and 5 cm of salt marsh substrate.

USA.) for plant material, P fertiliser and lime amendment treatments as fixed factors, and blocks as random factors. Normality and homogeneity of variance were tested and variance was modelled with GROUP statement of the function REPEATED when homogeneity was not respected. Degrees of freedom were adjusted accordingly. Significant differences between treatments were determined using protected LSD (LS means). Aikaike Information Criterion (AIC) was used to determine best models while Shapiro-Wilk and Kolmogorov-Smirnov tests were used to verify normality assumption.

Liming experiment in greenhouse

Concurrently with the field experiments, a liming experiment was conducted in the greenhouse to verify the effects of three doses of dolomitic lime on transplant establishment and seed germination of *C. paleacea* and *S. pectinata*.

Experimental design

The effects of lime doses (0, 2.5 and 7.5 kg m⁻³) were tested on two plant species in a completely randomised block design repeated five times. Blocking was used to take into account microenvironment variability (light and relative humidity, for example) within the greenhouse. Each experimental unit consisted of one container which measured 37 cm (length) × 56 cm (width) × 35 cm (height). The containers (30 in total) were filled with 8 cm of horticultural sand (at the bottom) and 20 cm of seawater-contaminated peat which had been adjusted to a final salinity (2.5‰) that was

representative of soil salinity in the seawater-contaminated bogs. At the beginning of the experiment, containers were watered (with rainwater) every two or three days. Frequency of watering was reduced over time to mimic water table levels observed in the field experiment throughout the 2011 growing season.

Plant material

Transplants of *C. paleacea* and *S. pectinata* and their seeds were collected from the Shippagan salt marsh in October 2011. Plugs were stored with humid peat at 4 °C, in the dark for three months, to break dormancy. Seeds were stored in the same conditions, but they were transferred into brackish water (16 ‰) for *S. pectinata* and 8 ‰ for *C. paleacea* two weeks before they were moved to the greenhouse, to break dormancy and improve germination rate (Wijte & Gallagher 1996, Van Der Valk *et al.* 1999). Lemma and palea of *S. pectinata* seeds and scales of *C. paleacea* seeds were removed by hand before soaking in brackish water (Stalter 1972, Walsh 1990). Six plants were transplanted along with two groups of 50 seeds in each experimental unit. Each group of seeds was constrained by a plastic mesh border 5 cm high and 10 cm in diameter to prevent dispersion (see Figure 2).

Liming treatments

Dolomitic lime (0, 2.5 and 7.5 kg m⁻³) was incorporated into the peat 2.5 months before the beginning of the experiment. Peat was regularly mixed and pH was measured every two weeks until the pH stabilised at 3.76 ± 0.04, 4.70 ± 0.13 and 6.22 ± 0.09, respectively.

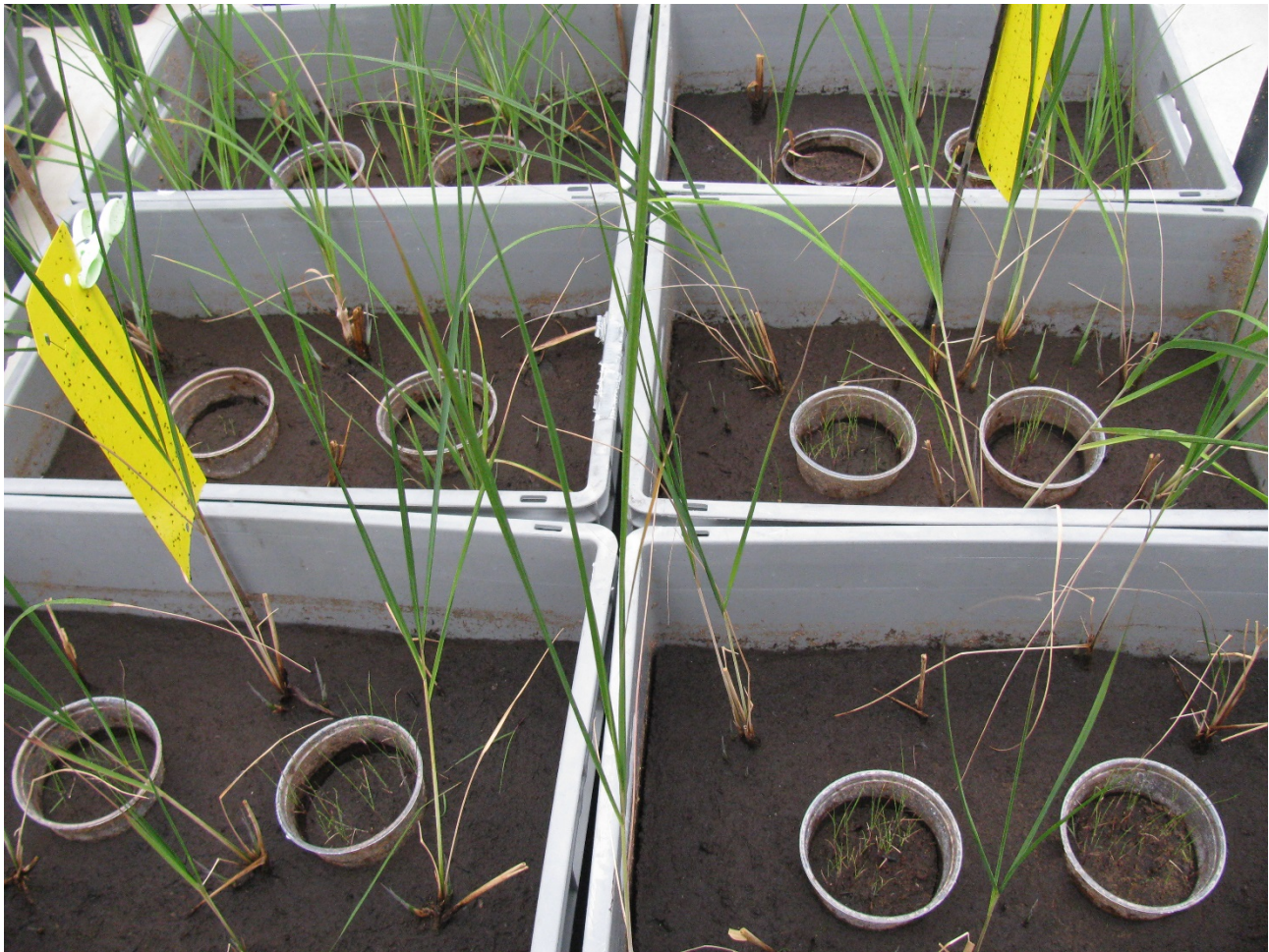


Figure 2. Photograph of one of the five blocks of the greenhouse experiment, comprising six experimental units. Each batch of 50 seeds is surrounded by a plastic mesh border.

Monitoring

Transplants were grown for a period of four months while seed germination was followed for two months. Soil characteristics (salinity, pH and redox) and biotic data (plant height and number of germinated seeds) were recorded every two weeks. At the end of the experiment, peat and leaf samples were collected from the transplants in each experimental unit of three representative blocks for chemical analysis. All aerial biomass was also collected from each container, but underground biomass was collected from two randomly chosen quarters *per* container. Chemical analyses employed the same techniques as described above for the field experiment.

Data analyses

Due to major differences in *S. pectinata* and *C. paleacea* morphology, data from the transplants were analysed separately for the two species. However, their seed germination rates were compared across treatments. Data analysis software

and procedures were the same as for the field experiment, i.e. using the MIXED procedure of SAS with the liming treatment as a fixed factor and blocks as random factors.

RESULTS

Field experiment

The main factor that influenced plant biomass and cover was the addition of P (Table A2); a summary of its effects is presented in Table 2. Phosphorous fertiliser had a positive effect on vegetation establishment, in that it improved total plant cover and spontaneous vegetation cover in all vegetation transfer treatments (Table 2 and Table A2). Introduced vegetation also tended to exhibit slightly greater cover in fertilised than in non-fertilised plots, but differences were only significant in the *C. paleacea* plots (Figure 3; significant interaction Vegetation type \times P addition; Table A2). Introduced,

Table 2. The positive effect of P addition on plant height, different types of aerial biomass and vegetation cover, and the mean foliar P concentrations and N/P quotients for *C. paleacea* and *S. pectinata* (SE in parentheses). Only variables for which there were significant differences (Table A2) are presented. For height, $n = 16$; for foliar nutrients, $n = 32$; for aerial biomass and cover, $n = 80$.

Plant response	With P	Without P
<i>Spartina pectinata</i> height (cm)	25 (8)	19 (7)
<i>Carex paleacea</i> height (cm)	36 (1)	31 (3)
Aerial biomass of spontaneous vegetation (g m^{-2})	116 (20)	62 (11)
Aerial biomass of introduced vegetation (g m^{-2})	45 (11)	23 (7)
Total aerial biomass (g m^{-2})	161 (13)	86 (23)
Spontaneous vegetation cover (%)	19 (3)	11 (2)
Total vegetation cover (%)	25 (3)	13 (2)
P concentration (mg g^{-1} dry mass)	1.6 (0.2)	1.0 (0.2)
N/P quotient	14.5 (1.4)	24.7 (1.7)

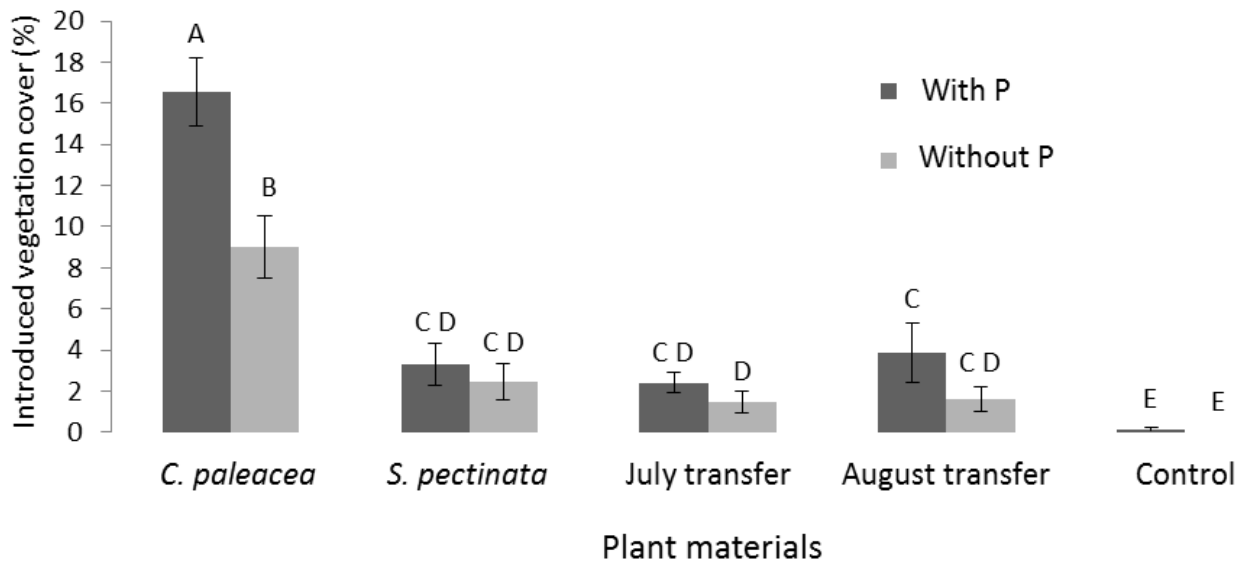


Figure 3. Mean percent cover of introduced vegetation according to the type of salt marsh plant material transferred and phosphorus fertiliser additions (error bars: \pm SE). *Carex paleacea* and *S. pectinata* were transplanted at the beginning of July 2011, whereas salt marsh diaspores were spread in mid-July and mid-August 2011. Plant cover was recorded in mid-summer 2012. Different capital letters indicate significant differences between treatments following protected LSD. ANOVAS for factorial block design on introduced vegetation cover: vegetation type ($F_{4,57} = 46$, $p < 0.001$), phosphorus ($F_{1,57} = 16$, $p = 0.004$), interaction ($F_{4,57} = 3.7$, $p = 0.019$). Lime treatment did not significantly affect introduced vegetation cover, nor did it interact significantly with the other treatments.

spontaneous and total aerial biomasses were higher in fertilised than in non-fertilised plots, reflecting the differences in plot cover. The height of *S. pectinata* and *C. paleacea* increased by 132 % and 116 %. However, underground biomass did not seem to be influenced by P addition (Table A2). In general, P was well assimilated by plants, as shown by the higher P concentration in plant tissue and lower N/P quotient in leaves of plants that received P amendment (Table 2, Table A3).

The addition of lime increased the peat pH from 3.5 ± 0.1 to 3.8 ± 0.1 (Lime effect: $F_{1,57} = 8.9$; $p = 0.004$), and doubled Ca concentration in peat (1.8 ± 0.2 versus 3.7 ± 0.7 mg g⁻¹; Lime effect $F_{1,57} = 6.5$, $p = 0.025$). Lime was also well absorbed by the plants, which exhibited a 170 % increase in Ca concentration compared to the plants that did not receive any (1.61 ± 0.38 versus 2.69 ± 0.39 mg g⁻¹; Table A3). The addition of lime also induced higher foliar Ca/Mg quotients than in plots with no addition of lime (1.31 ± 0.19 versus 0.87 ± 0.09). However, addition of lime reduced the aerial biomass of the introduced plant material in all vegetation treatments (26 ± 7 g m⁻² versus 42 ± 12 g m⁻²; Table A2). This was particularly obvious in the plots where *C. paleacea* had been introduced (significant interaction Vegetation type \times P addition; Table A2). All plots where *C. paleacea* had been introduced in the absence of lime exhibited the highest aerial biomass (159 ± 37.7 g m⁻²; Table A2).

The introduction of vegetation, either by transplant or salt marsh diaspore transfer, did not

significantly affect total vegetation cover or biomass (Table A2). However, plant composition differed greatly between treatments. Biomass of salt marsh species was highest in plots where *C. paleacea* had been transplanted (Figure 4) and this treatment also produced more cover than all other plant material treatments (Figure 3). Spontaneous vegetation biomass was lowest in the plots with transplants, and highest in control plots and in plots where plant material from salt marsh had been transferred (Table A2, Figure 4). Spontaneous vegetation is mainly composed of one species, *Juncus bufonius* L. Very few plant species apart from the one that was planted were found in transplant plots (data not shown). Salt marsh diaspore transfers led to considerably greater species richness. After two growing seasons, eleven typical salt marsh species successfully established in plots where the salt marsh diaspore transfer had been carried out, but their cover remained very low (Table A1).

Greenhouse liming experiment

Lime addition negatively affected the growth of transplants of *C. paleacea* and *S. pectinata*, (Table A4). Height and aerial biomass of *Carex paleacea*, and aerial and total biomass of *S. pectinata* decreased significantly as the dose of added dolomitic lime increased (Figure 5). The germination rate of *S. pectinata* seeds (24 ± 2.2 %) was five times that of *C. paleacea* seeds (5.1 ± 0.5 %), but no significant effect of lime addition was observed for germination of either species (Table A4).

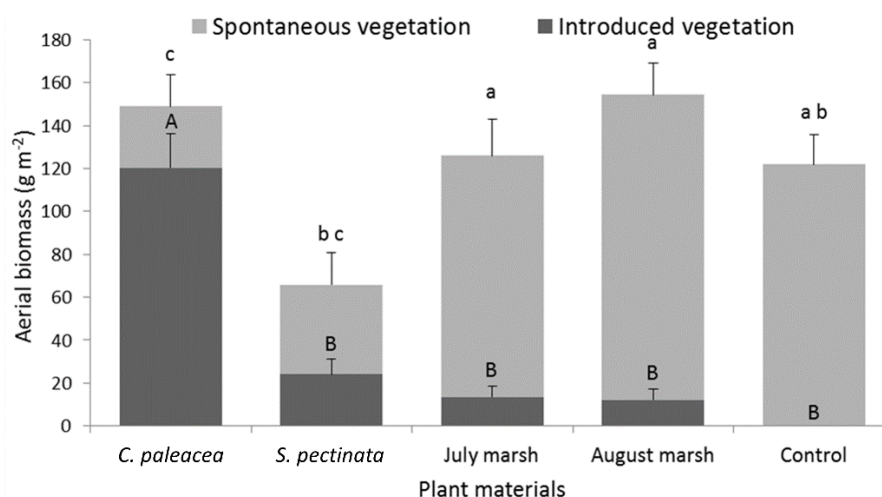


Figure 4. Mean aerial biomass of introduced and spontaneous vegetation according to the type of plant material introduced in the field experiment (pooled data across fertilisation treatment; error bars: \pm SE). Different letters indicate significant differences between treatments. Capital letters and lower error bars refer to the introduced vegetation while lowercase letters and upper error bars refer to spontaneous vegetation. See Table A2 for results of the statistical analyses. Note that total biomass (spontaneous + introduced vegetation) did not differ significantly between treatments.

Lime amendments increased the pH of the peat as predicted, but also reduced redox potential (Eh) in the root zone (Figure 6, Table A4). Lime also succeeded in raising the concentrations of exchangeable Ca and Mg in the peat but reduced extractable Al (Table 3). At the same time, Mn concentrations fell by 40 % and Fe concentrations decreased by 98 % (Table 3, Table A4). The concentration of exchangeable P did

not significantly increase following lime addition (lime treatment $F_{2,8} = 2.6$ and $p = 0.109$ for *C. paleacea* and *S. pectinata* pooled together) and P concentration was even lower in *C. paleacea* leaves from limed plots than from control plots (Table 3, Table A4). As a result, lime doubled the very low N/P quotient in leaves of *C. paleacea* (Table 3, Table A4).

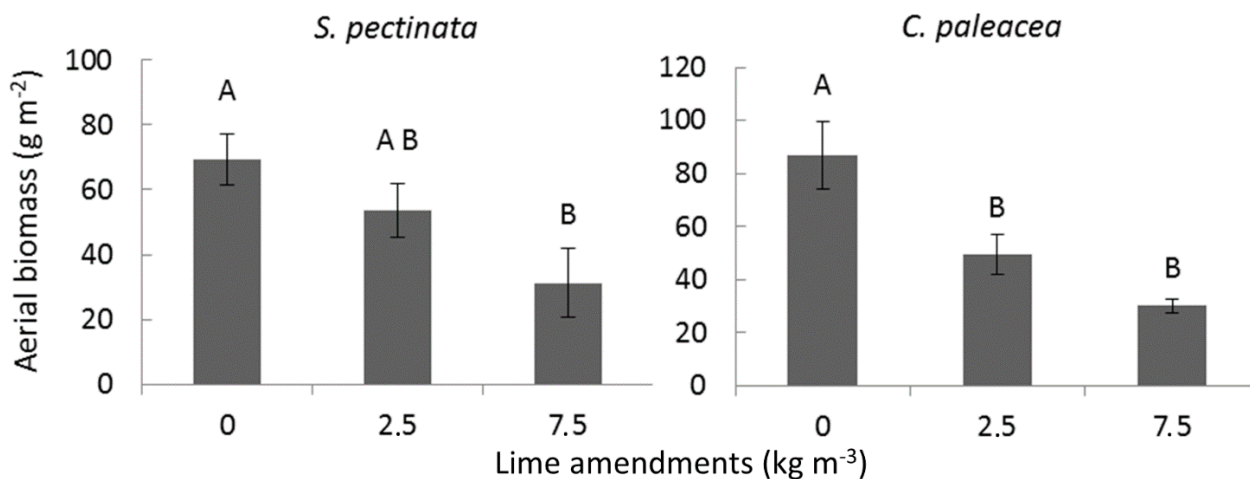


Figure 5. Effects of dolomitic lime treatments on *S. pectinata* and *C. paleacea* aerial biomass after four months of growth in the greenhouse (mean \pm SE) ($n = 15$). Different letters indicate significant differences following protected LSD. See Table A4 for results of the statistical analyses.

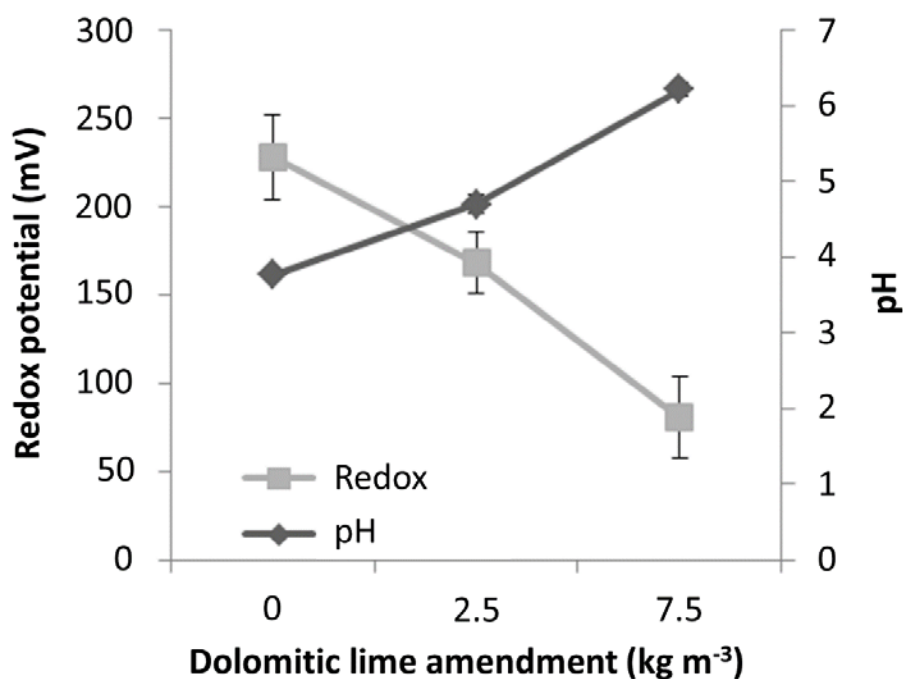


Figure 6. Effects of dolomitic lime treatments on the redox potential and pH in peat surrounding the root zone (mean \pm SE) in the greenhouse. Soil data for both species (*C. paleacea* and *S. pectinata*) are included in the means ($n = 30$). See Table A4 for results of the statistical analyses.

Table 3. The effects of lime additions on peat (soil exchangeable) and leaf nutrient concentrations in *C. paleacea* and *S. pectinata* plots in the greenhouse experiment. Only variables for which there were significant differences are presented. Mean ($n = 9$) concentrations and (SE) are given in mg g^{-1} for Ca, Mg and P or in $\mu\text{g g}^{-1}$ for Fe, Mn and Al.

Lime (kg)	Peat nutrient concentrations					Leaf nutrients	
	[Ca]	[Mg]	[Fe]	[Mn]	[Al]	[P]	[N/P]
<i>C. paleacea</i>							
0.0	1.2 (0.1)	1.4 (0.1)	109 (11)	23 (3)	235 (3)	1.5 (0.2)	9 (2)
2.5	6.7 (0.3)	4.9 (0.2)	29 (3)	27 (2)	51 (9)	1.0 (0.1)	18 (1)
7.5	11.9 (0.8)	8.3 (0.3)	1.0 (0.3)	15 (1)	19 (7)	1.0 (0.1)	20 (3)
<i>S. pectinata</i>							
0.0	1.2 (0.1)	1.6 (0.1)	82 (11)	26 (3)	228 (3)	1.8 (0.3)	9 (3)
2.5	6.7 (1.0)	5.0 (0.6)	41 (14)	25 (3)	61 (22)	1.3 (0.1)	16 (1)
7.5	12.1 (0.5)	8.6 (0.3)	2.0 (0.1)	15 (3)	12 (1)	1.2 (0.1)	16 (2)

DISCUSSION

Phosphorus fertilisation

The present study confirms that the addition of phosphorus helps plants adapt to the high salinity levels caused by seawater contamination of cutover bogs. The positive effect of P is indicated by changes in the leaf N/P quotient. A value above 16 indicates P limitation whereas a value below 14 indicates N limitation (Meuleman 2010). In the field experiment, N/P was 14.5 in the presence of P and close to 25 without P, demonstrating strong P limitation in peatlands contaminated by seawater. The addition of rock phosphate has also been found to promote the establishment of vascular plants in extracted bogs that are not contaminated by salt (Ferland & Rochefort 1997) and the establishment of *Polytrichum strictum* (Sottocornola *et al.* 2007) and other bryophytes in degraded blanket bog (O'Toole & Synnott 1971). In salt marsh restoration, P fertiliser is sometimes applied, but N fertilisation is predominant (Zedler 1984, Broome *et al.* 1988). In the case of seawater-contaminated cutover bogs, our results indicate that an application of P fertiliser is strongly recommended to improve the establishment of salt marsh plants.

Lime amendment

Liming did not improve plant growth. While the addition of lime did result in an increase in soil pH, it was expected that this would have resulted in improved nutrient availability while reducing Al availability in the soil (Griffin 1971, Ryan & Smillie 1975, Haynes 1982, Brady & Weil 2003). The addition of large amounts of lime to waterlogged acid soils was also expected to stimulate the precipitation of Al^{3+} in hydroxyl-Al polymers, which creates new adsorption surfaces for phosphates in the soil (Haynes 1982). This study shows that P availability in peat did not improve following lime addition and P concentration in leaves was even 1.5 times lower in heavily limed plots than in control plots (greenhouse experiment). Results similar to ours have been reported in different types of acid soils, where low soil pH was raised by the addition of lime, but P availability nevertheless decreased (Amarasiri & Olsen 1973, Sumner 1979). Furthermore, salt marsh plants did not seem to take advantage of improved Ca and Mg availability following liming. In the greenhouse experiment, the lime treatment dramatically reduced the availability in the peat of micronutrients such as Fe and Mn, which are important cofactors for enzyme activation and

chlorophyll synthesis in plants (Raven *et al.* 2003). Their lower concentrations in limed plots could be responsible for the poor growth observed in the greenhouse experiment, in association with the lower P absorption observed in the presence of lime.

Another factor that could explain the poor growth of *C. paleacea* and *S. pectinata* in limed substrates is the production of sulphide, which is toxic to plants (Lamers *et al.* 2013). Seawater is much richer in sulphate than freshwater ecosystems (Marschner 1995). Low pH (as in bogs) limits the activity of sulphate reducing bacteria (Hao *et al.* 1996); but lime additions, by increasing soil pH, might have improved the activity of bacteria including sulphate reducing bacteria. Indeed, the inverse relationship between redox potential and level of lime amendment suggests that bacterial activity did increase with soil pH. The stimulated consumption of oxygen might then have favoured the activity of anaerobic species such as sulphate reducing bacteria. The soil in the greenhouse was warmer than that in the field, which may also have increased microbial activity. Furthermore, liming reduced Fe availability (Table 3), thus reducing the amount of pyrite (FeS₂) that could be formed (Lamers *et al.* 2013), which would leave more sulphate to be reduced to sulphide by sulphate reducing bacteria. The reduction in phosphorus availability under liming conditions could also be related to increased bacterial activity, as bacteria can sequester significant amounts of nutrients (Richardson & Simpson 2011). However, *Spartina* spp. are known to be more tolerant than *Carex* spp. to sulphide (Lamers *et al.* 1998), which suggests either that other factors besides sulphide production influenced growth of these two species, or that *C. paleacea* is in fact more tolerant to sulphide than other *Carex* species.

Introduction of salt marsh vegetation

The higher cover of introduced plants and higher aerial biomass found in *C. paleacea* plots confirmed that this species is well adapted to the seawater-contaminated peatland environment. The slower response of *S. pectinata* is probably due to differences in growth rates between species. Spreading salt marsh diaspores requires less resources than transplanting individual plants and offers the advantage of improving species diversity in the restored site while not adversely affecting regeneration capacity in the donor site (personal observations, data not shown). Transferred diaspores can germinate, establish and grow in seawater-contaminated cutover bogs, but plant cover in the diaspore transfer plots was lower than in transplanted plots. This is probably related to the greater integrity

of transplants compared to rototilled salt marsh diaspores. Nevertheless, plant cover remained low even in the best treatments, leaving room for spontaneous vegetation growth. The high water level conditions during transplantation of *C. paleacea* and *S. pectinata* in June 2011 may have limited fast spontaneous vegetation establishment in the transplantation plots. The disturbance created by planters was much greater in the soft substrate than in the firmer substrate that was present during application of the other treatments (salt marsh diaspores transfer in July and August and control treatments). It is also likely that roots were disturbed while the seeds were being buried.

CONCLUSIONS

This study showed that rock phosphate addition improves the establishment of all plant material treatments trialled in this study by providing P, which is limiting for salt marsh vegetation in bogs. Transplantation of *C. paleacea* is recommended because it led to higher introduced vegetation cover and aerial biomass, whereas salt marsh diaspore transfer is recommended to promote species diversity. The latter technique is also more suitable for rehabilitation of large areas. Liming of seawater-contaminated and waterlogged peat substrates is not recommended because it appears to induce P, trace element and oxygen deficiencies, while potentially inducing the production of sulphide which is toxic to many plant species. N fertilisers could be tested in conjunction with P fertilisation to avoid inducing N limited plant growth. However, the effects of nutrient leaching on surrounding coastal habitats should be investigated if large areas are to be fertilised, to avoid potential eutrophication problems.

ACKNOWLEDGEMENTS

Financial support for this study was provided by the Natural Sciences and Engineering Research Council of Canada (NSERC), the Canadian Sphagnum Peat Moss Association (CSPMA) and SunGro Horticulture Ltd., within the program of the NSERC Industrial Research Chair in Peatland Management (Dr. Line Rochefort). The authors also wish to thank the Natural Resource Department of New Brunswick and Laval University. This project would not have been possible without the valuable help of field assistants and the PERG team. A special 'thank you' goes to the Coastal Zone Research Institute for access to their facilities during the field season.

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- Submitted 29 Oct 2015, revision 20 Jun 2016
Editor: Ab Grootjans
-

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Appendix



Figure A1. Bare peat in a sector of Pokesudie cutover bog in June 2011, eleven years after salt contamination.



Figure A2. Aerial photographs of the study sites, Pokesudie bog (left) and Shippagan bog (right). Both are located on the Acadian Peninsula of New Brunswick, Canada.

Table A1. Plant species diversity found in natural salt marshes one year after salt marsh diaspores were collected (15 species) was higher than in the experimental plots at the introduction site (11 species). Species are listed in descending order of cover in bog marsh transfer plots (mean \pm SE).

Plant species	Percent cover (%)	
	Bog marsh transfer plots (introduction site)	Natural salt marshes (donor site)
<i>Lysimachia maritima</i> (L.) Gal., Banf. & Sold.	2 \pm 1	11 \pm 2
<i>Juncus gerardii</i> Loisel.	2 \pm 0.5	21 \pm 2
<i>Juncus balticus</i> Willd.	2 \pm 0.5	5 \pm 2
<i>Carex paleacea</i> Shreb. Ex Wahl.	2 \pm 0.5	5 \pm 1
<i>Triglochin maritima</i> L.	1 \pm 0.5	4 \pm 1
<i>Plantago maritima</i> L.	1 \pm 0.5	4 \pm 1
<i>Potentilla anserina</i> L.	1 \pm 0.5	0.5 \pm 0
<i>Spartina</i> sp.	1 \pm 0.5	14 \pm 2
<i>Halerpestes cymbalaria</i> (Pursh) Greene	1 \pm 0.5	2 \pm 0.5
<i>Atriplex prostrata</i> Boucher ex de Candolle	0.5 \pm 0	1 \pm 0.5
<i>Festuca rubra</i> L.	0.5 \pm 0	1 \pm 0
<i>Limonium carolinianum</i> (Walter) Britton		3 \pm 1
<i>Salicornia depressa</i> Standley		3 \pm 0.5
<i>Carex Mackenziei</i> Krecz.		4 \pm 0
<i>Solidago sempervirens</i> L.		10 \pm 0
<i>Juncus bufonius</i> L.**	21 \pm 4	
<i>Eriophorum angustifolium</i> Honck.**	0.5 \pm 0	

** Spontaneous vegetation.

Table A2. Results of 3-way ANOVA testing for soil amendments and introduced plant material effects on biomass in the field experiment. Significant *p* values are in bold type (*n* = 80). Veg = the types of plant material introduced.

Sources	d.f.	Introduced ¹ veg aerial biomass		Spontaneous ² veg aerial biomass		Total aerial biomass		Underground biomass		Total biomass		Introduced veg cover		Spontaneous veg cover		Total cover	
		<i>F</i>	<i>p</i>	<i>F</i>	<i>p</i>	<i>F</i>	<i>p</i>	<i>F</i>	<i>p</i>	<i>F</i>	<i>p</i>	<i>F</i>	<i>p</i>	<i>F</i>	<i>p</i>	<i>F</i>	<i>p</i>
Blocks	3																
Veg	4	30.9	<0.001	3.5	0.026	0.8	0.596	1.1	0.381	2.2	0.099	46	<0.001	7.7	<0.001	1.6	0.200
Lime	1	4.3	0.043	0.3	0.610	0.02	0.892	0.3	0.565	0.2	0.645	0.0	0.973	0.02	0.883	0.2	0.676
Phosphorus	1	7.5	0.008	4.8	0.040	8.74	0.010	1.9	0.177	8.3	0.006	16	<0.001	10.8	0.003	21	<0.001
Veg × Lime	4	4.2	0.048	0.2	0.952	0.3	0.889	1.9	0.146	0.1	0.996	1.7	0.193	0.4	0.822	0.2	0.938
Veg × Phosphorus	4	1.3	0.281	1.6	0.208	0.9	0.562	1.0	0.446	0.3	0.866	3.7	0.019	1.4	0.277	0.7	0.620
Lime × Phosphorus	1	1.9	0.171	1.3	0.264	0.5	0.506	1.6	0.962	0.8	0.367	0.2	0.641	0.6	0.465	0.5	0.471
Veg × Lime × Phosphorus	4	1.6	0.179	0.7	0.608	0.4	0.796	0.2	0.962	0.9	0.485	1.0	0.437	0.8	0.552	0.4	0.783
Error	57																
Total	79																

¹ Vegetation from the salt marshes which had been transplanted or spread in the salt-contaminated cutover bogs is called “introduced vegetation”.

² Vegetation that grew spontaneously in the salt-contaminated cutover bogs is called “spontaneous vegetation”, and mainly comprised *Juncus bufonius* and *Eriophorum angustifolium*.

Table A3. Results of ANOVA testing for amendments and introduced plant material effects on leaf nutrient status in the field experiment. Foliar nutrients were analysed only in transplanted *C. paleacea* and *S. pectinata*, not in the other three vegetation treatments. Significant *p* values are shown in **bold** type (*n* = 40).

Sources	d.f.	Foliar nutrients							
		Phosphorus (P)		Calcium (Ca)		N/P		Ca/Mg	
		<i>F</i>	<i>p</i>	<i>F</i>	<i>p</i>	<i>F</i>	<i>p</i>	<i>F</i>	<i>p</i>
Blocks	3								
Veg	1	13	0.002	1.1	0.317	1.3	0.270	2.2	0.156
Lime	1	1.9	0.188	6.4	0.022	0.8	0.398	9.1	0.007
Phosphorus	1	19	0.001	0	0.953	20	0.004	0.5	0.511
Veg × Lime	1	0.1	0.779	0.5	0.513	0	0.841	1.2	0.285
Veg × Phosphorus	1	3.1	0.096	0	0.858	0.1	0.744	3.3	0.083
Lime × Phosphorus	1	0.8	0.372	0.2	0.650	0.7	0.411	0.8	0.391
Veg × Lime × Phosphorus	1	0.8	0.399	1.1	0.320	0	0.951	0.8	0.377
Error	21								
Total	31								

Table A4. Results of ANOVA testing for the effect of liming on peat chemical characteristics, plant characteristics, peat nutrients and leaf nutrients in the greenhouse experiment. Only nutrients for which statistically significant differences were observed are presented. *C. paleacea* and *S. pectinata* were not directly compared because their morphologies are different. Significant *p* values are shown in **bold** type.

		<i>C. paleacea</i>		<i>S. pectinata</i>	
		<i>F</i>	<i>p</i>	<i>F</i>	<i>p</i>
Peat characteristics¹	pH	442	<0.001	296	<0.001
	Redox	13	0.003	14	0.001
Plant characteristics¹	Germination rates	0.46	0.640	0.14	0.872
	Height of shoots	1.1	0.394	8.2	0.006
	Height of plants	6.0	0.025	0.1	0.953
	Aerial biomass	11	0.017	4.5	0.035
	Belowground biomass	0.1	0.912	3.8	0.075
	Total biomass	1.4	0.281	28	0.003
Peat nutrients²	Calcium (Ca)	151	0.002	69	<0.001
	Magnesium (Mg)	437	<0.001	88	0.005
	Iron (Fe)	67	0.001	8.6	0.018
	Manganese (Mn)	9.7	0.029	3.5	0.099
	Aluminium (Al)	359	<0.001	23	0.002
Leaf nutrients²	Phosphorus (P)	8.6	0.036	2.3	0.182
	N/P quotient	16	0.013	3.0	0.123

¹ All five blocks were analysed. Degrees of freedom: Blocks: 4; Lime: 2; Error: 8; Total: 14.

² Three blocks out of five were analysed. Degrees of freedom: Blocks: 2; Lime: 2; Error: 4; Total: 8.