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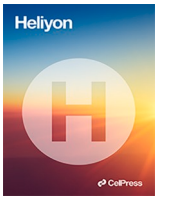

Comparative flooding tolerance of Typha latifolia and Phalaris arundinacea in wetland restoration: Insights from photosynthetic CO_2 response curves, photobiology and biomass allocation☆

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ABSTRACT

Tall helophytes such as *Typha latifolia* and *Phalaris arundinacea* often rapidly colonise after rewetting of former agricultural soil and are therefore often the first plants to contribute to the soil carbon pool. In this study we carried out a mesocosm experiment where these two species grew at three different water levels relative to the soil surface (−15 cm, 0 cm, +15 cm). After eight weeks' growth, measurements of photosynthetic CO_2-response curves, stomatal conductance and chlorophyll fluorescence of photosystem II were carried out to detect flooding stress. After 10 weeks' growth, the plants were harvested and biomass production, biomass allocation and specific leaf area were determined. *T. latifolia* had a higher and more stable photosynthetic performance across all water level treatments, which resulted in an overall higher aboveground and belowground production than *P. arundinacea*. In contrast, V_{cmax} and J_{max} decreased by 41 % and 42 %, respectively from drained to flooded conditions with signs of flooding stress as impairment of the photosynthetic apparatus. Moreover, increasing water level resulted in maintenance of aboveground organs for *P. arundinacea* but a decrease in allocation to belowground organs. *P. arundinacea* did not invest in a higher specific leaf area to counter the decreased photosynthesis under flooding. From −15 cm to 0 cm water levels, *P. arundinacea* showed a 68 % reduction in belowground biomass, which has negative implication for carbon retention immediately after rewetting. In contrast, recolonization of *T. latifolia* is likely to be a suitable contributor to the soil carbon pool due to its stable physiology and high above- and belowground biomass production at all water depths, and also likely under natural water level fluctuations. We showed that even though both species are generally considered wetland plants, they are likely to support considerably different photosynthetic carbon assimilation and soil carbon sequestration rates.

1. 1. introduction

Draining of peat wetlands for agricultural purposes has for centuries caused increased CO_2 emissions from the soil to the

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atmosphere. Peatland restoration on agricultural soil is an important action to substantially reduce global CO_2 emissions from soil to the atmosphere, with potential GHG emission reductions of 0.31–3.38 Gt CO_2-equivalents [1]. Rewetting aims to re-establish anaerobic soil conditions, restoring the natural wetland biogeochemistry, but cannot usually restore the natural ecosystem and its functions immediately [2,3]. When rewetting of a former wetland occurs, this starts a natural successional process towards paludification [4]. However, in the early successional stages after rewetting, wetlands often lack the peat-forming community characteristic of infertile, natural peatlands [5,6]. Instead, community composition is generally dominated by tall graminoids, also termed helophytes [3,7–9]. This helophytisation of rewetted sites, caused mainly by elevated nutrient availability in degraded former agricultural soil, has implications beyond biodiversity. A difference in species composition can alter litter quality and consumption and production of carbon in the soil and rhizosphere [10]. Moreover, the aerenchymatous tissue of tall graminoid species is highly conductive for gases, which can either increase or decrease methane emissions from wetland sites, depending on the balance between internal CH_4 transport and rhizosphere oxidation [11–13].

Two helophyte species that often rapidly colonise and dominate species composition in northern Europe after peatland rewetting are T*ypha latifolia* L. (Typhaceae) and *Phalaris arundinacea* L. (Poaceae) [9]. These two tall graminoid species are both highly productive and well-adapted to waterlogged and anoxic soils, due to their aerenchyma and efficient O_2 transport from the atmosphere to below-ground organs [14,15]. However, although both species have the very high percentage volume of aerenchyma typical of wetland taxa [16], they differ in their aeration mechanisms. The two gas transport mechanisms in wetland plants are 1) simple molecular diffusion, and 2) pressurized convective gas flow. Gas transport in *P. arundinacea* occurs by simple molecular diffusion (H. Brix unpublished data). Here, the respiratory uptake of oxygen by the below-ground tissue creates a concentration gradient that drives oxygen diffusion from the atmosphere to the rhizomes and roots [17]. In contrast, for *T. latifolia* pressurized convective flow of gases is the most important mechanism that drives internal oxygen transport [18]. Plants with convective flow have a throughflow of air such that when pressurization occurs in one part of the plant, a bulk flow of internal gases is driven towards a venting point elsewhere in the gas transport system. The differences in internal pressure are generated by temperature and water vapour pressure gradients between internal gas spaces and the atmosphere [19,20]. This adaptation greatly increases the oxygen flux through the rhizomes and indirectly to roots, allowing plants to grow in deeper water [21,22].

How these first recolonisers perform after rewetting is important, as they play a key role in initial biomass production and carbon accumulation in the soil [23]. However, although wetland plants are adapted to wet and flooded conditions, they can still suffer oxygen deprivation during long periods of flooding [24]. When oxygen availability becomes restricted in flooded and waterlogged soils, aerobic rhizome and root respiration can be inhibited, which results in shortage of ATP and lowered root activity [25]. Glycolysis and fermentation can maintain production of energy production during hypoxia, but such anaerobic respiration can lead to build up of compounds such as ethanol and lactic acid, which are toxic [26]. Flooding can also lead to increased stomatal closure and decreased stomatal conductance, decreasing CO_2 uptake, resulting in lower photosynthetic carbon assimilation [27]. Moreover, flooding can also cause damage to photosystem II (PSII), resulting in less photochemical energy captured and hence lower fluorescence yield of green leaves [28]. This may coincide with down-regulation of electron transport, but rather than occurring due to photodamage, a lower electron transport rate can also occur as a photoprotective mechanism to avoid reactive oxygen species (ROS) by dissipation of energy [29]. Plants that experience flooding for shorter or longer periods can therefore suffer impaired photosynthetic apparatus, lower biomass production and changes in biomass allocation, which can negatively affect the efficiency of soil carbon burial in wetlands [30].

Recolonization by tall helophytes after wetland rewetting is likely to support considerably different carbon sequestration rates and carbon cycling compared with natural wetlands. How these rapid recolonisers perform physiologically and in biomass accumulation needs to be investigated, to determine to what extent they can support recovery of wetland ecosystem functioning and soil carbon accumulation in the initial successional stage after rewetting. In this study, we investigated the photosynthetic responses and growth of these two highly productive wetland species, T. *latifolia* and *P. arundinacea,* to three different water regimes (drained, waterlogged and flooded). We used CO_2 response (A/C_i) curves and chlorophyll fluorescence to determine photosynthetic performance and physiological stress. We hypothesized that even though both species are well-adapted for wetland habitats and both tolerate long periods of flooding, *T. latifolia* would perform better in deeper water due to its pressurized gas transport [22].

2. Materials and methods

2.1. Plant material and experimental setup

Single shoots of *T. latifolia* including ~10 cm long rhizomes were collected in late May 2021 from a small pond in East Jutland, Denmark (56°24′45.6″N 10°09′37.3″E). Similar *P. arundinacea* individuals were collected in late May from an artificial shallow lake, Årslev Engsø, in East Jutland, Denmark (56°08′04.9″N 10°02′31.4″E). At the Påskehøjgård research garden near Aarhus, Denmark (56°13′45.5″N, 10°07′33.7″E), *P. arundinacea* shoots were laid horizontally in shallow water in the greenhouse, until adventitious shoots and roots were produced from the stem nodes. Shoots and rhizomes of *T. latifolia* were planted in pots and placed outdoors in large water trays. The mean temperature was 17.4 ± 3.7 °C over the ~3-week cultivation period. Approximately 20 cm–40 cm tall newly developed shoots of *P. arundinacea* and 30 cm–50 cm tall shoots of *T. latifolia* were then planted in 3.5 L pots with holes in the bottom containing 50 % commercial growth substrate and 50 % sand. Replicates of both species were placed in ten 70.5 cm × 70.5 cm × 59.5 cm tubs (L × W × H). Upon starting the experiment, the pots with plants were placed in a waterlogged condition in the tubs (i.e., with the water level reaching the top of the pots) to ensure that the plants had the same baseline before the onset of the experiment. After being waterlogged for four days, one pot with either *T. latifolia* or *P. arundinacea* was placed at a drained (−15 cm), waterlogged

(0 cm) or flooded (+15 cm) position in each of ten tubs. The experimental setup was located in a greenhouse with temperature logged hourly (TG 4100, Tinytag Plus 2, Gemini Data Loggers, Chichester, UK) and irradiance logged every 10 min (Odessey Photosynthetic Active Radiation Logger, Dataflow Systems Ltd, Christchurch, NZ). The mean temperature in the greenhouse during the whole experiment was 24.2 ± 5.7 °C, while mean daytime irradiance was 218 ± 263 μmol photons m^{-2} s^{-1} with a maximum irradiance of 1085 μmol photons m^{-2} s^{-1} at a height of approximately 59.9 cm (the top edge of the tubs and initial canopy height). Water temperature was logged twice daily at 06.00 h and 18.00 h (TG-4100, Tinytag Aquatic 2, Gemini Data Loggers, Chichester, UK) in each of the tubs, and the mean water temperature during the experiment was 22.5 ± 1.9 °C.

Each pot was fertilized with 500 mL of a solution of macro- and micronutrients twice weekly. The full-strength nitrogen and phosphorus macronutrient solution (Pioner NPK Makro, Azelis, Kongens Lyngby, DK) contained NO_3–N (119.0 mg L^{-1}), NH_4–N (74.0 mg L^{-1}) and P (23.0 mg L^{-1}). Additional macronutrient concentrations were K (154.0 mg L^{-1}), Mg (30.0 mg L^{-1}) and S (39.0 mg L^{-1}). The micronutrient solution consisted of Pioner Mikro Plus Iron (0.1 mL L^{-1}; Azelis, Kongens Lyngby, DK) and also contained 1.61 % Fe, 0.25 % B, 0.13 Cu, 0.63 % Mn, 0.06 % Mo, 0.31 % Zn. Additionally, the micronutrient solution consisted of an iron chelate 6 % EDDMA (0.08 g L^{-1}; Azelis, Kongens Lyngby, DK). The relatively strong macronutrient solution was chosen to ensure that the plants were not limited by any interaction between nutrient availability and flooding stress [31]. Before fertilizing, each replicate was removed from the tubs and then fertilized to ensure that the whole pot was saturated. After a waiting period of 3–5 min, the replicates were returned to their correct position in the tubs.

The experiment had the two fixed main factors "Species" (*T. latifolia* vs. *P. arundinacea*) and "Water level" (−15 cm, 0 cm, +15 cm) resulting in a 2 × 3 factorial setup. Nine replicates of *T. latifolia* survived at 0 cm and −15 cm, and 10 replicates of *P. arundinacea* survived at −15 cm and +15 cm. Seven replicates of *T. latifolia* and 9 replicates of *P. arundinacea* survived at +15 cm and 0 cm, respectively. The unbalanced number of replicates was due to loss of some replicates well into the experimental period, and all dead plants were excluded from the analysis. All physiological measurements were carried out in the greenhouse.

2.2. Chlorophyll fluorescence measurements

After six weeks' growth in the greenhouse, variable chlorophyll fluorescence measurements were carried out with a portable fluorometer (Mini-PAM II fluorometer, Heinz Walz GmbH, Effeltrich, Germany). One young but fully developed leaf was chosen on each of the replicates and marked with a small aluminum tag at the base of the leaves for identification. Φ_{PSII} (operational yield of Photosystem II under the prevailing irradiance) measurements were initiated at 06:00 h and were afterwards conducted every other hour on the same leaf for each replicate. The final measurement was completed at 04:00 h the next day.

2.3. Photosynthetic A/C_i curves

After eight weeks' growth, gas exchange measurements were carried out using an infrared gas analyzer (LI-COR 6800 Portable Photosynthesis System, LI-COR, Lincoln, NE, USA). For *T. latifolia*, each measurement was performed on one young fully developed leaf per replicate, and the gas analyzer's leaf cuvette was placed ~20 cm down the leaf from the leaf tip. For *P. arundinacea*, measurements were done on the third or fourth fully-developed leaf from the shoot tip. Leaf chamber settings during all gas exchange measurements were a constant temperature of 28 °C to mimic the relatively high temperatures in the greenhouse, a relative humidity of 65 %, constant air flow of 500 μmol s^{-1}, a fan speed of 10,000 rpm, and a constant light intensity in the leaf chamber of 1500 μmol photons m^{-2} s^{-1}.

When the gas exchange rate was stable at a reference CO_2 concentration of 400 μmol mol^{-1}, leaf area was specified on the instrument and an A/C_i curve auto program was launched. For each replicate, the A/C_i curve followed a sequence of CO_2 concentrations of 400, 300, 200, 100, 50, 0, 400, 400, 500, 600, 800, 1000, 1200, 1400, 1600, 1600, 2000, and 400 μmol mol^{-1}. The Excel fitting tool developed by Sharkey (2016) [32] for C3 plants was used to fit the CO_2 response curves and to estimate key physiological parameters. The estimated cardinal points from each curve replicate were the maximum carboxylation rate of Rubisco (V_{cmax}), maximum electron transport rate (J_{max}), CO_2-compensation point (Γ), and triose phosphate utilization (TPU).

2.4. Biomass

Ten weeks after the onset of the experiment and after the completion of physiological measurements, each replicate was harvested and separated into leaf, stem, dead biomass and belowground biomass fractions. Soil was carefully removed from the plant material, which was then rinsed until there was no remaining substrate. The biomass fractions were dried in an oven at 70 °C for 72 h to a constant dry weight and placed in an exicator before weighing. Leaf area was obtained by scanning all living leaves in a LI-COR LI-3100 Area Meter (LI-COR inc., Lincoln, NE, USA) and from this specific leaf area $\left(\boldsymbol{SLA} = \frac{\boldsymbol{Leaf\ area\ (m^2)}}{\boldsymbol{leaf\ biomass\ (kg)}}\right)$ was calculated. From the weighted biomass fractions a range of biomass parameters were calculated: total biomass (total biomass = leaf biomass + stem biomass + root biomass + dead biomass), leaf mass ratio $\left(\boldsymbol{LMR} = \frac{\boldsymbol{Leaf\ biomass\ (g\ DW)}}{\boldsymbol{Total\ biomass\ (g\ DW)}}\right)$, stem mass ratio $\left(\boldsymbol{SMR} = \frac{\boldsymbol{Stem\ biomass\ (g\ DW)}}{\boldsymbol{Total\ biomass\ (g\ DW)}}\right)$, root mass ratio $\left(\boldsymbol{RMR} = \frac{\boldsymbol{Root\ biomass\ (g\ DW)}}{\boldsymbol{Total\ biomass\ (g\ DW)}}\right)$ and shoot:root ratio $\left(\boldsymbol{Shoot:root\ ratio} = \frac{\boldsymbol{Leaf\ biomass\ (g\ DW) + Stem\ biomass\ (g\ DW) + Dead\ biomass\ (g\ DW)}}{\boldsymbol{Root\ biomass\ (g\ DW)}}\right)$.

2.5. Statistics

All data analyses were carried out using the software R Studio (R Studio 2021.09.0, Boston, MA, USA). A two factorial ANOVA (Analysis of Variance) was performed with a type III sum of squares for all biomass and photosynthetic parameters. The two categorical factors were species (*T. latifolia* and *P. arundinacea*) and water level (Drained (−15 cm), Waterlogged (0 cm), Flooded (+15 cm)) and the ANOVA analysis included the interaction term. A Tukey's HSD *post hoc* test was performed that compared all means on a 5 % significant level. Homogeneity of variances was tested on all data using Levene's test. In cases where homogeneity of variances was not achieved, the data were either log- or square root transformed.

For the 24-h measurements of Φ_{PSII} a two way repeated measures ANOVA (RMANOVA) was performed with a type III sum of squares. For each of the two species, the two categorical factors were water level (Drained (−15 cm), Waterlogged (0 cm), Flooded (+15 cm)) and time (continues measurements every other hour) with the RMANOVA analysis including the interaction term. A Tukey's HSD *post hoc* test was performed that compared all means on a 5 % significant level. Levene's test was used to test homogeneity of variance, and a power transformation was performed when homogeneity of variance was not achieved.

3. Results

3.1. Biomass

Overall, *T. latifolia* produced more total biomass, for all biomass fractions, than *P. arundinacea*. The exception was stem biomass, which was higher for *P. arundinacea* in drained conditions than for *T. latifolia* (Table 2). Despite the generally high biomass production for *T. latifolia*, the total biomass from drained to waterlogged was reduced by 36 % (Table 2). However, the total biomass in flooded conditions did not differ from either drained or waterlogged conditions. *P. arundinacea* generally responded negatively to increased water level, emphasized by a significant interaction for all biomass fractions, and produced more biomass in drained conditions (Tables 1 and 2). Compared to *P. arundinacea,* biomass fractions for *T. latifolia* did not show any particularly strong response to increasing water level, however belowground biomass for *T. latifolia* in waterlogged conditions and *P. arundinacea* in drained conditions were comparable (Table 2).

Biomass allocation to the leaves was not affected by differences in water level in either species, however LMR was higher for *T. latifolia* as indicated by a significant effect of species (Table 1, Fig. 1a). Stem allocation was significantly higher for *P. arundinacea*, while flooding in *P. arundinacea* resulted in higher stem allocation compared to the two other treatments. There was no difference in stem allocation for *T. latifolia* across all treatments (Fig. 1b). Water level did not affect root allocation in *T. latifolia* (Fig. 1c); in contrast, in the deeper water levels of the waterlogged and drained conditions, RMR for *P. arundinacea* decreased, corresponding to a significant interaction (Table 1, Fig. 1c). In drained conditions, the species had similar RMR, however, again *P. arundinacea* was affected by flooding as reflected by the increase in shoot:root mass ratio, which corresponded to an increase in SMR and a decrease in RMR with increased water level (Fig. 1d). Shoot:root mass ratio in *T. latifolia* was not affected by water level (Fig. 1d).

3.2. SLA and stomata conductance

SLA was more than twice as high in *P. arundinacea* and there was no significant effect of water level for either species (Tables 1 and 3). For stomatal conductance there was also no effect of water level, but stomatal conductance was twice as high for *T. latifolia* than *P. arundinacea,* as shown by a significant species effect (Tables 1 and 3).

Table 1
F-ratios from the two-way ANOVA illustrating the effects of the main factors species (*Typha latifolia*, *Phalaris arundinacea*) and water level (drained, waterlogged, flooded) and their interaction on biomass fractions, leaf mass ratio (LMR), shoot mass ratio (SMR), root mass ratio (RMR), shoot:root mass ratio, specific leaf area (SLA), Stomatal conductance (g_s) maximum carboxylation rate of Rubisco (V_{cmax}), maximum electron transport rate (J_{max}), and CO_2 compensation point (Γ). * P-value <0.05; ** <0.01; *** <0.001.

	Main factor		Interaction
	Species	*Water level*	*Species × Water level*
Leaf biomass	28.7***	18.5***	7.6**
Stem biomass	1.9	12.1***	3.2*
Dead biomass	48.9***	6.6**	3.9*
Belowground biomass	15.2***	33.7***	13.3***
Total biomass	29.3***	11.8***	3.4*
LMR	51.2***	1.8	1.6
SMR	223.5***	9.5***	4.3*
RMR	1.4	23.4***	11.8***
Shoot:root mass ratio	2.0	20.6***	9.9***
SLA	60.6***	5.3	3.9
g_s	20.2***	3.6	2.7
V_{cmax}	59.9***	7.1**	4.5**
J_{max}	58.5***	8.1***	4.4*
Γ	1.2	2.9	1.5

Table 2
Total biomass and biomass fractions; biomass of leaves, stems, dead plant litter and belowground biomass of *Typha latifolia* and *Phalaris arundinacea* (mean ± SD) grown at the three different water levels (drained, waterlogged, flooded). Different letters indicate significant differences between treatments.

Biomass fraction (g)	Typha latifolia			Phalaris arundinacea		
	Drained	Waterlogged	Flooded	Drained	Waterlogged	Flooded
Total biomass	76.01 ± 12.63^{a}	48.11 ± 18.85^{bc}	61.07 ± 23.56^{ab}	31.55 ± 13.72^{cd}	15.85 ± 5.83^{de}	9.92 ± 9.00^{e}
Leaf biomass	34.15 ± 7.12^{a}	23.41 ± 10.09^{a}	30.46 ± 12.61^{a}	8.43 ± 3.31^{b}	4.10 ± 1.69^{b}	2.62 ± 3.10^{c}
Stem biomass	8.07 ± 2.42^{ab}	5.58 ± 2.63^{bc}	6.70 ± 2.33^{abc}	10.7 ± 4.39^{a}	5.33 ± 2.14^{bc}	4.43 ± 3.32^{c}
Dead biomass	7.80 ± 1.39^{a}	4.30 ± 1.97^{b}	4.34 ± 1.43^{b}	2.28 ± 1.19^{c}	2.09 ± 1.30^{cd}	0.87 ± 0.45^{d}
Belowground biomass	25.99 ± 6.83^{a}	14.81 ± 6.26^{ab}	20.81 ± 7.98^{a}	10.10 ± 5.67^{b}	3.73 ± 1.31^{c}	2.00 ± 2.82^{d}

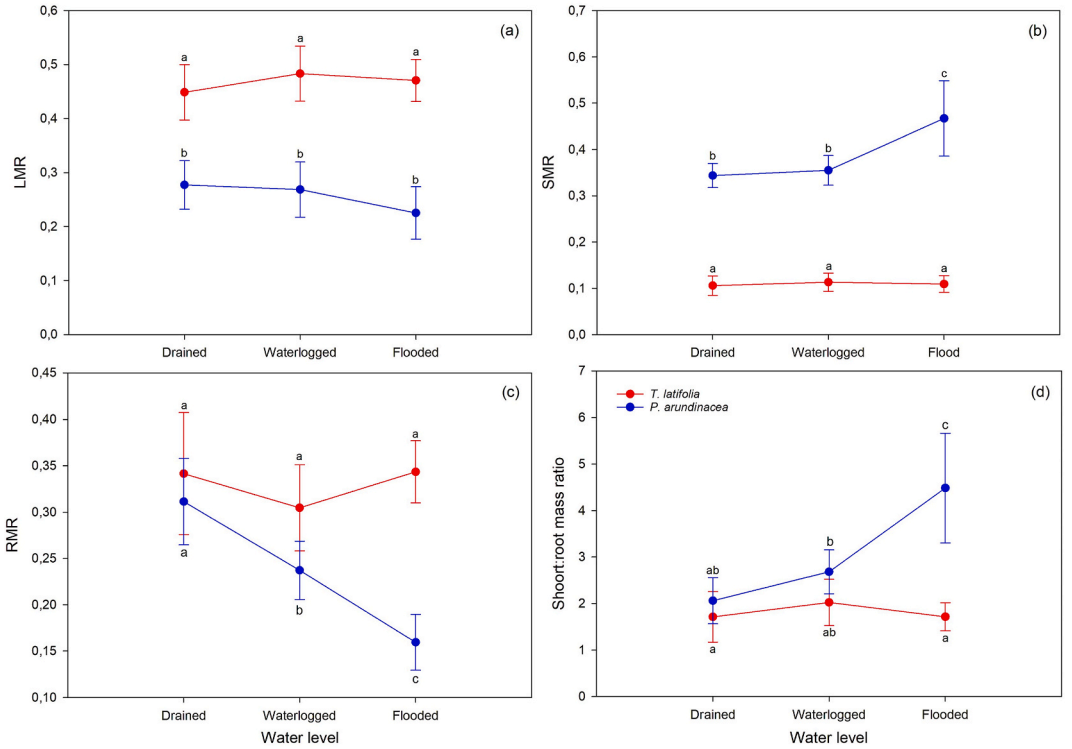

Fig. 1. Leaf mass ratio, LMR (a), stem mass ratio, SMR (b), root mass ratio, RMR (c) and shoot:root mass ratio (d) (mean ± SD) of *Typha latifolia* and *Phalaris arundinacea* after 10 weeks of growing at different water levels (drained, waterlogged, flooded). Different letters indicate significant differences between treatments.

Table 3
Specific leaf area (SLA), stomatal conductance (g_s), maximum carboxylation rate of RUBISCO (V_{cmax}), maximum electron transport rate (J_{max}) and CO_2 compensation point (Γ) of *Typha latifolia* and *Phalaris arundinacea* (mean ± SD) grown at the three different water levels (drained, waterlogged, flooded). Different letters indicate significant treatments between treatments.

Parameter	Typha latifolia			Phalaris arundinacea		
	Drained	Waterlogged	Flooded	Drained	Waterlogged	Flooded
SLA (m^{-2} kg^{-1})	12.23 ± 1.54^{a}	13.83 ± 2.21a	13.80 ± 1.64a	29.18 ± 3.74^{b}	30.24 ± 7.49 b	23.38 ± 7 .48^{b}
g_s (mol m^{-2} s^{-1})	0.66 ± 0.09^{a}	0,63 ± 0.16a	0.69 ± 0.11a	0.41 ± 0.10^{b}	0.32 ± 0.11 b	0.26 ± 0.09^{b}
V_{cmax} (μmol m^{-2} s^{-1})	101.73 ± 7.26^{a}	98.95 ± 14.39a	104.15 ± 10.64a	52.80 ± 9.18^{b}	43.25 ± 14.82BCE	31.12 ± 15.15^{c}
J_{max} (μmol m^{-2} s^{-1})	194.22 ± 9.01^{a}	185.18 ± 26.77a	194.42 ± 20.24a	102.99 ± 17.78^{b}	84.43 ± 27.32BCE	59.57 ± 30.39^{c}
Γ (μmol mol^{-1})	54.41 ± 2.71^{a}	54.11 ± 4.31a	54.09 ± 3.21a	59.63 ± 7.64^{ab}	63.95 ± 12.36 ab	70.00 ± 15.59^{b}

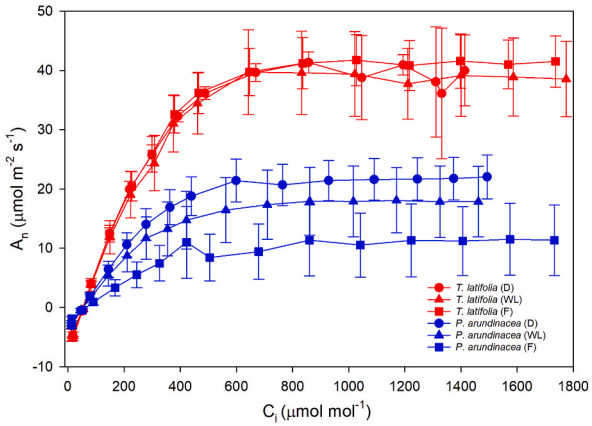

Fig. 2. CO_2-response curves (mean ± SD) of *Typha latifolia* and *Phalaris arundinacea* after eight weeks of growing at three different water levels (D: Drained, WL: Waterlogged, F: Flooded).

3.3. A-C_i curves and cardinal points

Overall, *T. latifolia* had higher photosynthetic rates than *P. arundinacea* (Table 3, Fig. 2). Photosynthesis in *T. latifolia* was unaffected by water level with no differences in cardinal point values between treatments (Table 3). However, the carboxylation rate of Rubisco (V_{cmax}) and the electron transport rate (J_{max}) for *P. arundinacea* were negatively affected by increased water level, indicated by a significant interaction between species and water level (Tables 1 and 2). V_{cmax} and J_{max} decreased by 41 % and 42 %, respectively, from drained to flooded conditions. In drained conditions, V_{cmax} for *T. latifolia* was 48 % higher than for *P. arundinacea*, while in flooded conditions it was 70 % higher for *T. latifolia* than for *P. arundinacea.* For the CO_2-compensation point (Γ), there was no significant effect of species and water level, however, Γ was generally higher for *P. arundinacea* (Tables 1 and 3).

3.4. Fluorescence measurements

Diel variation in Φ_{PSII} was significant for both species, with lower Φ_{PSII} during the day at high light intensities (Table 4, Fig. 3a and b). Water level had no effect on Φ_{PSII} for *T. latifolia* (Table 4). Significantly lower values of Φ_{PSII} (indicated by letters from Tukey HSD *posthoc* test, Fig. 3b) for the flooded *P. arundinacea* replicates during night (no light) were detected, which was emphasized by a significant interaction (Table 4).

4. Discussion

Our study showed that even though both species are specialized wetland plants with well-developed aerenchyma [16], they differed strongly in their response to increased water level. *T. latifolia* had a stable response to the different water regimes and was unaffected by either the drained or flooded conditions in terms of biomass yield, biomass allocation and physiology. On the other hand, *P. arundinacea* responded negatively to increasing water level, resulting in reduced biomass yield, changes in allocation patterns and lowered photosynthetic rates. Due to differences in aeration mechanisms of the two species, where *T. latifolia* is supported by pressurized flow and *P. arundinacea* is restricted to simple molecular diffusion, we anticipated that *T. latifolia* would be less affected by different flooding regimes than *P. arundinacea.* Based on the results from this study, we suggest that the large differences in the overall performance between the two plant species can be primarily explained by their different aeration mechanisms. *T. latifolia* is recognized as an obligate wetland species [14] that thrives in flooded conditions which is sustained by its pressurized flow, increasing oxygen availability in rhizomes and roots. In this experiment, growth and photosynthesis performance (both V_{cmax} and J_{max}) for *T. latifolia* were stable among water level treatments. However, though *T. latifolia* is considered a wetland obligate, this result suggests that *T. latifolia* can sustain high production and biomass accumulation also during drier summer periods where the water table is well below the soil surface. Li et al. (2004) [33] showed that a change from flooding to periodic drought resulted in a reduction in both photosynthetic performance and growth, indicating that *T. latifolia* is susceptible to drought. The drained treatment in our experiment did

Table 4
F-ratios from the two-way repeated measures ANOVA (RMANOVA) illustrating the effects of the main factors Water level (drained, waterlogged, flooded) and Time and their interaction on Yield of Photosystem II (PSII). * P-value <0.05; ** <0.01; *** <0.001.

	Main factor		Interaction
	Water level	*Time*	*Water level × Time*
Typha latifolia	1.0	235.5***	0.3
Phalaris arundinacea	32.6***	210.6***	1.9*

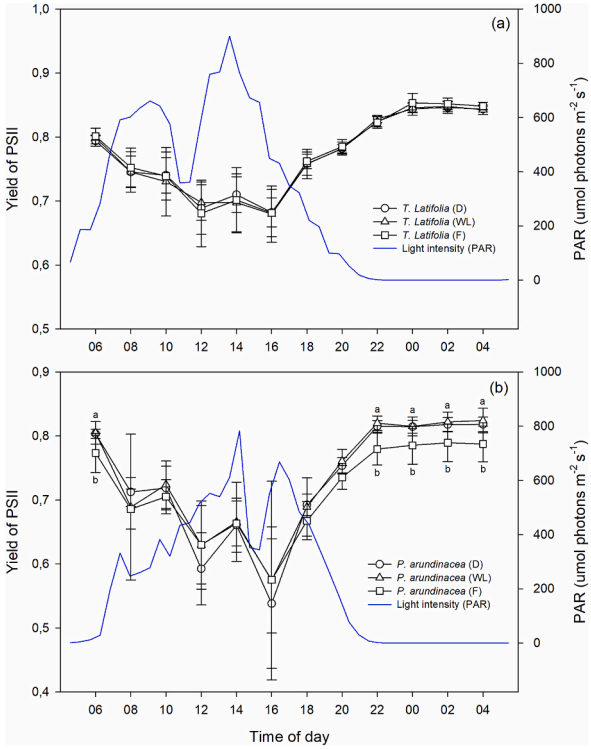

Fig. 3. Diurnal variation in Yield of PSII (mean ± SD) with corresponding light intensities for *Typha latifolia* (a) and *Phalaris arundinacea* (b) after six weeks of growing at different water levels (D: Drained, WL: Waterlogged, F: Flooded) Letters in (b) indicate significant differences between Drained/Waterlogged treatments and Flooded treatments. The remaining letters are not shown for clarity.

not represent drought since there was still a water supply from −15 cm below the soil surface. However, it is very likely that even shorter periods with drought would be detrimental for *T. latifolia*. The negative response to increased water level in *P. arundinacea*, a facultative wetland plant [14], is most likely due to the combined effect of oxygen deficiency due to the slow diffusion rate in water and insufficient diffusive oxygen flux in the aerenchyma [34].

The flooding stress experienced by *P. arundinacea* in the flooded treatment was apparent when looking at its physiological and photosynthetic performance. The carboxylation rate of Rubisco (V_{cmax}) in *P. arundinacea* significantly decreased with increased flooding, which is likely caused by the energy crisis associated with reduced root respiration resulting in reduced active nitrogen uptake [35]. *P. arundinacea* can take up and use both NO_3^- and NH_4^+, however, due to the lower energy cost of ammonium uptake and the possible reduced NO_3^- availability, sustained NH_4^+ uptake could have caused additional stress by cytoplasmic acidosis [36,37]. The reduced internal nitrogen is thought to both reduce the quantity of the Rubisco enzyme, causing an overall decreased Rubisco activity resulting in lower CO_2 assimilation rates [27,38]. Moreover, C3 plants like *P. arundinacea* invest a larger fraction of nitrogen in Rubisco compared to C4 plants, meaning C3 photosynthesis can be more negatively affected during nitrogen deficiency [39]. Regarding stomatal closure caused by high water level, flooding creates oxygen deprivation in belowground parts, resulting in transport of ABA to shoots, causing stomatal closure [40]. In *P. arundinacea* this could have contributed to the decreased carboxylation rates in flooded replicates as stomatal conductance decreased [41]. The low photosynthesis rates must therefore mainly have been caused by reduced Rubisco activity. Moreover, specific leaf area (SLA) was stable for both species across all water levels. SLA typically increases when leaves develop under water, since this trait reduces the diffusion resistance for gases, and as a consequence increases the rates of CO_2 entry for photosynthesis [42]. It is likely that the more efficient gas transport and aeration in *T. latifolia* during flooding permits high and stable photosynthesis rates, which makes investments in higher specific leaf area redundant for this species when flooded. On the other hand, *P. arundinacea* does not invest in a higher specific leaf area to counter the decreased photosynthesis under flooding, and thus seems not to have this acclimation capacity. Moreover, the significantly decreased fluorescence yield of photosystem II in dark-adapted (during night hours) *P. arundinacea* that experienced flooding indicates chronic stress due to flooding [43], however, there was no clear sign of leaf senescence. The detected reduced fluorescence yield results from loss of light energy through energy dissipation due to impairment of the photosynthetic apparatus [44]. Whether this stress is caused by damage of the PSII reaction centers or a photoprotective mechanisms cannot be determined, since the energy dissipation from non-photochemical quenching was not measured directly. This flooding stress has likely impaired photochemical electron transfer and utilization of energy indicated by the significant reduction in the maximum electron transport rate (J_{max}) when comparing drained and flooded plants [45].

Overall, our data suggest that a prolonged raised water level above the soil surface causes both decreased carbon fixation rates and decreased light energy utilization in *P. arundinacea,* resulting in a dramatic reduction of photosynthesis and biomass production.

However, the physiological stress associated with flooding in our study is not entirely consistent with other studies on flooding tolerance in *P. arundinacea*. Many previous studies in North America have found similar flooding responses in *P. arundinacea* and *T. latifolia,* with almost identical biomass production and high yield in both species across a broad hydrological range [16,46]. As these studies have been carried out on North American genotypes, the inconsistency might be explained by higher genetic variability among these genotypes stemming from multiple introductions of *P. arundinacea* from different European locations to North America, resulting in hybridization events creating more genetic variability and phenotypic plasticity [47]. In contrast, although different genotypes from within Europe can significantly differ in both their morphology and physiology, in northern Europe, *P. arundinacea* is considered to prefer moderately wet soil where the mean water table is from 0 to 20 cm below ground level [48,49]. Interestingly, this corresponds with our finding that the belowground biomass production of *P. arundinacea* was significantly reduced by 63 % from −15 cm to 0 cm. This result indicates that prolonged periods with relatively small increases in water table level can substantially decrease the direct carbon accumulation by *P. arundinacea* in rewetted soils. Moreover, in a rewetting scenario the goal is often to increase carbon retention and halt CH_4 emissions [50]. This is accomplished by maintaining the water level close to the surface to retain an oxidized upper soil layer [51,52]. However, this is likely to result in unsatisfactory carbon inputs by *P. arundinacea*. The concurrent decrease in physiological performance in *P. arundinacea* with increasing water level also highlights the importance of including this dimension when investigating potential candidate species used in wetland restoration and paludiculture, because this allows researchers to identify optimal growing conditions, stress, and acclimation potential before the onset of more costly large-scale systems.

Not only were biomass production and yield negatively affected by flooding in *P. arundinacea*, but biomass allocation was also affected. Allocation to leaves was stable among treatments for both *T. latifolia* and *P. arundinacea,* which indicates that water level did not affect leaf allocation. Decreased leaf allocation is a rare response to flooding and is a more common response to nutrient limitation, low light and suboptimal temperatures [53]. A stable leaf allocation is an important component of wetland restoration and carbon management, because the production and decomposition depend on the quality of the organic material from different species [54]. Here, lignin content and high C/N ratios of aboveground plant litter are important for carbon retention in soil because they prolong the decomposition of the organic material. Aboveground parts of *T. latifolia* have a relatively high lignin content, which combined with a high seasonal aboveground biomass production makes it a suitable plant for wetland carbon sequestration due to low decomposition rates [55]. Moreover, the stable root allocation exhibited by *T. latifolia* supports this, as it produces a relatively high belowground biomass, which implies that *T. latifolia* can sustain a large direct contribution to the soil carbon pool even at different water levels and likely also at natural water level fluctuations in restored wetlands [56]. In contrast, *P. arundinacea* had stable leaf allocation (LMR) but with a substantial reduction in root allocation (RMR), which might be due to the combined effect of allocation to aboveground parts and a reduced root growth caused by the lowered oxygen availability in the underground parts. Waliszewska (2021) [57] showed that *P. arundinacea* had a relatively low lignin content (leaves and stems; 15.42 %) compared with a range of other grasses, with the wetland grass *Phragmites australis* (Cav.) Trin. ex Steud. having the highest lignin content (leaves and stems; 21.99 %) in their study. Moreover, *P. arundinacea* increased its stem allocation (SMR) corresponding to an increased shoot:root ratio when flooded, which is often a response to decreased light availability [58]. Early in the experiment, flooded *P. arundinacea* was most likely light-limited due to the deeper water. In general, this suggests that *P. arundinacea* escapes flooding stress via morphological changes, since it does not have the ability to upregulate its photosynthetic performance due to lack of plasticity, with its simple diffusion aeration mechanism being the physiological bottleneck. With the substantial decreases in total biomass and root allocation, and increases in shoot:root ratio with increased flooding suggest that *P. arundinacea* is not be the most suitable wetland helophyte species to contribute to the soil carbon pool in restored wetlands.

5. Conclusion

Our study shows that even though both *T. latifolia* and *P. arundinacea* are wetland plants, they differ greatly in their responses to extended periods of flooding. *T. latifolia* has a stable physiology and maintains high aboveground and belowground biomass production for all water regimes. In contrast, *P. arundinacea* is less able to acclimate sufficiently to waterlogged and flooded water regimes, which leads to stress response in the photosynthetic apparatus and a drastic reduction in biomass production, and particularly belowground biomass production. Therefore, recolonization of these species in restored wetlands could lead to significantly different organic carbon input to the rewetted soil, where *T. latifolia* has the biggest potential to reestablish the organic carbon accumulation function after rewetting due to its high biomass production and its ability to retain its production, even in periods of lower water table. Since our study only is applicable to responses in the initial first year after rewetting, studies carried out over longer periods are needed to determine whether the responses are sustained and the consequences for carbon accumulation. Future studies should also consider including physiological parameters to better determine optimal growth conditions for potential restoration plant species.

Data availability

Data will be made available on request. Data associated with this study have not been deposited into a publicly available repository.

CRediT authorship contribution statement

Asger Buur Jensen: Writing - review & editing, Writing - original draft, Visualization, Methodology, Formal analysis, Data curation, Conceptualization. **Franziska Eller:** Writing - review & editing, Visualization, Validation, Supervision, Resources, Methodology. **Brian K. Sorrell:** Writing - review & editing, Supervision, Funding acquisition, Conceptualization.

Declaration of competing interest

The authors declare the following financial interests/personal relationships which may be considered as potential competing interests.

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References

[1] J. Leifeld, L. Menichetti, The underappreciated potential of peatlands in global climate change mitigation strategies, Nat. Commun. 9 (2018) 1071.
[2] A. Günther, A. Barthelmes, V. Huth, H. Joosten, G. Jurasinski, F. Koebsch, J. Couwenberg, Prompt rewetting of drained peatlands reduces climate warming despite methane emissions, Nat. Commun. 11 (2020) 1644.
[3] J. Kreyling, F. Tanneberger, F. Jansen, S. van der Linden, C. Aggenbach, V. Blüml, J. Couwenberg, V.J. Emsens, H. Joosten, A. Klimkowska, W. Kotowski, L. Kozub, B. Lennartz, Y. Liczner, H. Liu, D. Michaelis, C. Oehmke, K. Parakenings, E. Pleyl, A. Poyda, S. Raabe, M. Röhl, K. Rücker, A. Schneider, J. Schrautzer, C. Schröder, F. Schug, E. Seeber, F. Thiel, S. Thiele, B. Tiemeyer, T. Timmermann, T. Urich, R. van Diggelen, K. Vegelin, E. Verbruggen, M. Wilmking, N. Wrage-Mönnig, L. Wołejko, D. Zak, G. Jurasinski, Rewetting does not return drained fen peatlands to their old selves, Nature communication 12 (2021) 5693.
[4] W.J. Mitsch, L. Zhang, K.C. Stefanik, A.M. Nahlik, C.J. Anderson, B. Bernal, M. Hernandez, K. Song, Creating wetlands: primary succesion, water quality changes, and self-design over 15 years, Bioscience 62 (2012) 237–250.
[5] J.E. Moeslund, D.K. Andersen, A.K. Brunbjerg, H.H. Bruun, C. Fløjgaard, S.N. McQueen, B. Nygaard, R. Ejrnæs, High nutrient loads hinder successful restoration of natural habitats in freshwater wetlands, Restor. Ecol. (2022) 1–13.
[6] A.J.P. Smolders, H.B.M. Tomassen, L.P.M. Lamers, B.P. Lomans, J.G.M. Roelofs, Peat bog formation restoration by floating raft formation: the effects of groundwater and peat quality, J. Appl. Ecol. 39 (2002) 391–401.
[7] S. Roth, T. Seeger, P. Poschlod, J. Pfadenhauer, M. Succow, Establishment of helophytes in the course of fen restoration, Appl. Veg. Sci. 2 (1999) 133–136.
[8] S.A. Kozlov, L. Lundin, N.A. Avetov, Revegetation dynamics after 15 years of rewetting in two extracted peatlands in Sweden, Mires Peat 18 (2016) 1–17.
[9] M. Chytry, J.H.J. Schaminee, EUNIS habitat classification: expert system, characteristic species combinations and distribution maps of European habitats, Appl. Veg. Sci. 23 (2020) 648–675.
[10] D. Zak, C. Roth, V. Unger, T. Holdhammer, N. Fenner, C. Freeman, G. Jurasinski, Unraveling the importance of polyphenols for microbial carbon mineralization in rewetted riparian peatlands, Front. Environ. Sci. 7 (2019) 147.
[11] H.J. Laanbrook, Methane emissions from natural wetlands: interplay between emergent macrophytes and soil microbial processes. A mini-review, Ann. Bot. 105 (2010) 141–153.
[12] D.M.D. Hendriks, J. van Huissteden, A.J. Dolman, Multi-technique assessment of spatial and temporal variability of methane fluxes in peat meadow, Agric. For. Meteorol. 150 (2010) 757–774.
[13] R.J.E. Vroom, F. Xie, J.J.M. Geurts, A. Chojnowska, A.J.P. Smolders, L.P.M. Lamers, C. Fritz, Typha latifolia paludiculture effectively improves water quality and reduces greenhouse gas emissions in rewetted peatlands, Ecol. Eng. 124 (2018) 88–98.
[14] M.S. Fennesy, J.K. Cronk, W.J. Mitsch, Macrophyte productivity and community development in created freshwater wetlands under experimental hydrological conditions, Ecol. Eng. 3 (1994) 469–484.
[15] T.D. Colmer, Long-distance transport of gases in plants: a perspective on internal aeration and radial oxygen loss from roots, Plant Cell Environ. 26 (2003) 17–36.
[16] S.M. Kercher, J.B. Zedler, Flood tolerance in wetland angiosperms: a comparison of invasive and noninvasive species, Aquat. Bot. 80 (2004) 89–102.
[17] W. Armstrong, Aeration in higher plants, Adv. Bot. Res. 7 (1979) 225–332.
[18] M. Bendix, T. Tornbjerg, H. Brix, Internal gas transport in Typha latifolia L. and Typha angustifolia L. 1. Humidity-induced pressurization and convective throughflow, Aquat. Bot. 49 (1994) 75–89.
[19] J. Armstrong, E. Armstrong, P.M. Beckett, Phragmites australis: venturi- and humidity induced pressure flows enhance rhizome aeration and rhizosphere oxidation, New Phytol. 120 (1992) 197–207.
[20] H. Brix, B.K. Sorrell, P.T. Orr, Internal pressurization and convective gas flow in some emergent freshwater macrophytes, Limnol. Oceanogr. 37 (1992) 1433, 1420.
[21] V. Vretare Strand, The influence of ventilation systems on water depth penetration of emergent macrophytes, Freshw. Biol. 47 (2002) 1097–1105.
[22] B.K. Sorrell, I. Hawes, Convective gas flow development and the maximum depths achieved by helophyte vegetation in lakes, Ann. Bot. 105 (2010) 165–174.
[23] D. Were, F. Kansiime, T. Fetahi, A. Cooper, Jjuunko, Carbon sequestration by wetlands: a critical review of enhancement measures for climate change mitigation, Earth Systems and Environment 3 (2) (2019) 327–340.
[24] M.L. Otte, What is stress to a wetland plant? Environ. Exp. Bot. 46 (2001) 195–202.
[25] H. van Veen, M. Akman, C.L. Jamar, D. Vreugdenhil, M. Kooiker, P. van Tienderen, L.A.C.J. Voesenek, M.E. Schranz, R. Sasidharan, Group VII ethylene response factor diversification and regulation in four species from flood-prone environments, Plant Cell Environ. 37 (2014) 2421–2432.
[26] J. Pan, R. Sharif, X. Xu, X. Chen, Mechanisms of waterlogging tolerance in plants: research progress and prospects, Front. Plant Sci. 11 (2021), 627331.
[27] C. Liao, C. Lin, Physiological adaption of crop plants to flooding stress, Proceedings of the National Science Council 25 (2001) 148–157.
[28] Z. Liu, R. Cheng, W. Xiao, Q.N. Wang, Effects of off-season flooding on growth, photosynthesis, carbohydrate partitioning, and nutrient uptake in Distylium chinense, PLoS One 9 (9) (2014), e107636.
[29] W.W. Adams, B. Demmig-Adams, A.S. Verhoeven, D.H. Barker, "Photoinhibition" during winter stress: involvement of sustained xanthophyll cycle-dependent energy dissipation, Aust. J. Plant Physiol. 22 (1994) 261–276.
[30] S.R. Pezeshki, Wetland plant responses to soil flooding, Environ. Exp. Bot. 46 (2001) 299–312.
[31] J.A. Nyman, Lindau, Nutrient availability and flooding stress interact to affect growth and mercury concentration in Taxodium distichum (L.) Rich. Seedlings, Environ. Exp. Bot. 125 (2016) 77–86.
[32] T.D. Sharkey, What gas exchange data can tell us about photosynthesis, Plant Cell Environ. 39 (2016) 1161–1163.
[33] S. Li, S.R. Pezeshki, S. Goodwin, Effects of soil moisture regimes on photosynthesis and growth in cattail (Typha latifolia), Acta Oecol. 25 (2004) 17–22.
[34] E.J.W. Visser, L.A.C.J. Voesenek, B.B. Vartapetian, M.B. Jackson, Flooding and plant growth, Ann. Bot. 91 (2003) 107–109.
[35] N.M. Crawford, D.M. Glass, Molecular and physiological aspects of nitrate uptake in plants, Trends Plant Sci. 3 (1998) 389–395.
[36] H. Brix, B. Lorenzen, J.T. Morris, H.-H. Schierup, B.K. Sorrell, Effects of Oxygen and Nitrate on Ammonium Uptake Kinetics and Adenylate Pools in Phalaris Arundinaceae L. And Glyceria maxima (Hartm.) Holmb 102B, Proceedings of the Royal Society of Edinburgh, 1994, pp. 333–342.
[37] D.L. Hao, J.Y. Zhou, S.Y. Yang, W. Qi, K.J. Yang, Y.H. Su, Function and regulation of ammonium transporters in plants, Int. J. Mol. Sci. 21 (2020) 3557.

[38] A. Herrera, Responses to flooding of plant water relations and leaf gas exchange in tropical trees of a black-water wetland, Front. Plant Sci. 4 (2013) 1–12.
[39] R.F. Sage, R.W. Pearcy, J.R. Seemann, The nitrogen use efficiency of C3 and C4 plants, Plant Physiology 85 (1987) 355–359.
[40] J. Rodríguez-Gamir, G. Ancillo, M.C. Gonzáles-Mas, E. Primo-Millo, D.J. Iglesias, M.A. Forner-Giner, Root signalling and modulation of stomatal closure in flooded citrus seedlings, Plant Physiol. Biochem. 49 (2011) 636–645.
[41] F.P.O. Mollard, G.G. Strikier, E.L. Ploschuk, P. Insausti, Subtle topographical differences along a floodplain promote different plant strategies among Paspalum dilatatum subspecies and populations, Austral Ecol. 35 (2010) 189–196.
[42] T.D. Colmer, L.A.C.J. Voesenek, Flooding tolerance: suites of plant traits in variable environments, Funct. Plant Biol. 36 (2009) 665–681.
[43] H.K. Lichtenthaler, U. Rinderle, The role of chlorophyll fluorescence in the detection of stress conditions in plants, Crit. Rev. Anal. Chem. 19 (1988) 29–85.
[44] M.D. Fernandez, Changes in photosynthesis and fluorescence in response to flooding in emerged and subemerged leaves of Pouteria orinicoensis, Photosynthetica 44 (2006) 32–38.
[45] B. Genty, J.M. Briantais, N.R. Baker, The relationship between the quantum yield of photosynthetic electron transport and quenching of chlorophyll fluorescence, Biochim. Biophys. Acta 990 (1989) 87–92.
[46] R.C. Miller, J.B. Zedler, Responses of native and invasive wetland plants to hydroperiod and water depth, Plant Ecol. 167 (2003) 57–69.
[47] J. Molofsky, A.R. Collins, E. Imbert, T. Bitinas, S. Lavergne, Are invasive genotypes superior? An experimental approach using native and invasive genotypes of invasive grass Phalaris arundinacea, Open J. Ecol. 7 (2017) 125–139.
[48] C. Brodersen, S. Lavergne, Molofsky, Genetic variation in photosynthetic characteristics among invasive and native populations of reed canary grass (Phalaris arundinacea), Biol. Invasions 10 (2008) 1317–1325.
[49] W. Wichtmann, C. Schröder, H. Joosten, Paludiculture – Productive Use of Wet Peatlands, Schweizerbart Science Publishers, Stuttgart, 2016.
[50] C.D. Evans, M. Peacock, A.J. Baird, R.R.E. Artz, A. Burden, N. Callaghan, P.J. Chapman, H.M. Cooper, M. Coyle, E. Craig, A. Cumming, S. Dixon, V. Gauci, R. P. Grayson, C. Helfter, C.M. Heppell, J. Holden, D.L. Jones, J. Kaduk, P. Levy, R. Matthew, N.P. McNamara, T. Misselbrook, S. Oakley, S.E. Page, M. Rayment, L. M. Ridley, K.M. Stanley, J.L. Williamson, F. Worral, R. Morrison, Overriding water table control on managed peatland greenhouse gas emissions, Nature 593 (2021) 548–552.
[51] D.M.D. Hendriks, J. van Huissteden, A.J. Dolman, M.K. van der Molen, The full greenhouse gas balance of an abandoned peat meadow, Biogeosciences 4 (2007) 411–424.
[52] V. Huth, A. Günther, G. Jurasinski, S. Glatzel, The effect of an exceptionally wet summer on methane effluxes from a 15-year re-wetted fen in north-east Germany, Mires Peat 13 (2013) 1–7.
[53] H. Poorter, K.J. Niklas, P.B. Reich, J. Oleksyn, P. Poot, L. Mommer, Biomass allocation to leaves, stems and roots: meta-analyses of interspecific variation and environmental control, New Phytol. 193 (2012) 30–50.
[54] C.C. Overbeek, S.F. Harpenslager, J.P. Zuidam, E.E. Loon, L.P.M. Lamers, M.B. Soons, W. Admiraal, J.T.A. Verhoeven, A.J.P. Smolders, J.G.M. Roelofs, H.G. van der Geest, Drivers of vegetation development, biomass production and the initiation of peat formation in a newly constructed wetland, Ecosystems 23 (2020) 1019–1036.
[55] A.P. Neiff, J.J. Neiff, S.L. Casco, Leaf litter decomposition in three wetland types of the Parana River floodplain, Wetlands 26 (2006) 558–566.
[56] L. Ren, F. Eller, C. Lambertini, W.Y. Guo, H. Brix, B.K. Sorrell, Assessing nutrient responses and biomass quality for selection of appropriate paludiculture crops, Sci. Total Environ. 664 (2019) 1150–1161.
[57] B. Waliszewska, M. Grzelak, E. Gawel, A. Spek-Dzweigala, A. Sieradzka, W. Czekala, Chemical Characteristics of selected grass species grass species from Polish meadows and their potential utilization for energy generation purposes, Energies 14 (2021) 1669.
[58] Y. Yang, D. Yu, Y. Li, Y. Xie, X. Geng, Phenotypic plasticity of two submerged plants in response to flooding, J. Freshw. Ecol. 19 (1) (2004) 69–76.