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1

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Combined effect of temporal inundation and above-ground-cutting on the growth of two emergent wetland plants, *Phragmites* and *Bolboschoenus* sp.

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Background. *Phragmites* are a common foundation species found in inland and brackish estuarine ecosystems. *Phragmites* offer a wide range of habitats for wetland organisms and provide essential functions, including nutrient cycling, pollutant filtration, wave energy reduction, and soil stabilization. However, excessive *Phragmites* growth can degrade the quality of wetland habitats, thereby reducing the functions of restored wetlands.

Methods. In this study, we examined the effectiveness of vegetation management techniques, such as aboveground cutting and temporal inundation with varying depth and periodicity, in controlling growth of *Phragmites* and adjacent vegetation. Differences in growth responses to manipulated inundation stress between *P. australis* and *B. planiculmis* were measured. **Results.** An inundation stress of 10-50cm caused significantly greater growth inhibition in *B. planiculmis* compared to *P. australis*. Combining aboveground-cutting and inundation treatments results in a significant restraining effect on the growth and survival rate of *P. australis*. The growth characteristics of *P. australis*, including stem volume and biomass, decreased, and its mortality rate increased. Our manipulated experiment suggests a combined treatment approach of moderate inundation, such as 5-10 cm for 20-30 days, and aboveground cutting to manage the overgrowth of *P. australis* in restored brackish wetlands. **Keywords** brackish wetland, food plant, mesocosm experiment, mortality, vegetation management, water level

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Abstract

Background. *Phragmites* are a common foundation species found in inland and brackish estuarine ecosystems. *Phragmites* offer a wide range of habitats for wetland organisms and provide essential functions, including nutrient cycling, pollutant filtration, wave energy reduction, and soil stabilization. However, excessive *Phragmites* growth can degrade the quality of wetland habitats, thereby reducing the functions of restored wetlands.

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Introduction

Over the past century, rapid land conversion, reclamation, and urban expansion have resulted in a significant loss of natural wetland habitats. Various types of wetland restoration procedures have been undertaken as mitigation strategies (Zedler and Kercher 2005). Nevertheless, it is frequently reported that even after restoration projects are carried out to enhance the degraded function and condition of wetland ecosystems, the desired wetland condition could not be maintained for the expected periods. This site management problem can occur when restoration plans do not adequately reflect core ecological processes (e.g., Gupta et al. 2020; Keddy 2010; Mitsch and Gosselink 2015; Thomaz 2023; van Biervliet et al. 2020; Wang et al. 2019; Zarekarizi et al. 2020) required to restore wetland functions, or when these functions are lost due to poor management and maintenance practices after restoration project. For example, restored wetlands may degrade rapidly due to changes in habitat structures, increased sedimentation rates resulting from an altered hydrological regime, or the expansion of invasive species. Changes in the physical habitat structure and hydrological regime of restored wetlands closely related to the level of species richness and the composition of plant communities (Wang et al. 2016). A deeper understanding of the key response traits of restored plant communities is critical for designing proper management measures to achieve long-term restoration objectives (An et al. 2022).

Phragmites australis (common reed) is an important foundation species that provides diverse ecological roles in estuarine ecosystems (Dolinar et al. 2016). *P. australis* tends to

establish clonal populations storing large amounts of plant biomass and providing complex microhabitats and food sources for local communities, supporting the regional food web structure. However, when the distribution of *P. australis* become rapidly expanding and unbalanced with other vegetation types, it can displace local endemic plant communities (e.g., *Bolboschoenus* sp.). This can result in a reduction of overall biodiversity and degradation of related ecosystem functions. For example, invasive genotypes of *Phragmites* have been observed in Europe and the United States. With their rapid spread and growth characteristics, unlike those of native *Phragmites* communities, they are changing the structure and function of natural and restored wetland ecosystems (Hazelton et al., 2014; van der Putten, 1997). Uncontrolled and rapid expansion of *Phragmites* communities can simplify the diversity of habitats in wetlands, such as the vegetation type, and reduce the richness of species that can thrive on wetland vegetation (Greet and King 2019).

The rapid expansion of *P. australis* in many reed-dominated wetlands poses a global management problem that scientists have tried to tackle with various methods, including physical removal (e.g., cutting or fire; Tanaka et al. 2016; Zengel et al. 2018), biological control (Blossey et al., 2020), chemical control (Schad et al. 2021), and water level control. While some strategies like fire or chemical control have been shown to effectively prevent further expansion of *P. australis* (Bonello and Judd, 2020), their use is limited in the field due to safety and chemical regulation concerns in protected wetlands (c.f., primary use of herbicide; Martin and Blossey, 2013). Reports of the remaining methods being tested have been inconsistent, and they can vary based on the wetland type of managed site, competing plant species, and environmental characteristics. Given this complexity outcomes, experts suggest that efficient management of *P. australis* requires the development of site-specific methods tailored to local environmental factors.

Phragmites beds in restored wetlands on Eulsuk Island provide supporting services to various wetland species, including the Oriental Great Reed Warbler (*Acrocephalus orientalis*), Leopard cat (*Prionailurus bengalensis*), Brown sesarmid crab (*Chiromantes dehaani*), and diverse insect species (Busan Metropolitan City 2022). However, the recent rapid expansion of *P. australis* accelerates ecological succession from wetland vegetation to terrestrial vegetation, simplifying habitat types in the restored wetland. Several management trials attempted to remove the overgrown *P. australis* using heavy machines, but they failed, spending a significant portion of the management cost (Busan Metropolitan City 2022). Continued expansion of *P. australis* can further threaten the loss of *B. planiculmis*, which is one of the primary food sources for migratory birds during the winter season. Given the current site condition, it is highly necessary to develop a sustainable way of controlling the expansion of *P. australis*.

In this study, we conducted manipulated plant growth experiments near the restored wetland site on Eulsuk Island. The pot experiments setup in the restored wetland evaluated the effects of two types of management measures (inundation and aboveground cutting) on two emergent wetland species. Wetland inundation (e.g., water level control) is a relatively easy-to-apply management measure in restored wetlands with floodgates installed. Aboveground

removal cannot be applied over large areas but they can limit the expansion of plant communities in a short term. Based on the manipulated experiments, an optimal management plan was proposed to most effectively apply the two methods (i.e., inundation and aboveground cutting) to control *P. australis* expansion in the restored wetlands.

Materials & Methods

Study site

The pot experimental setup and plant collection were conducted in the Nakdong River estuary (NRE). The study site has a humid subtropical climate based on the Köppen climate classification (Busan Metropolitan City 2022). The annual average temperature is 14.6°C. The annual rainfall is approximately 1,354 mm. Over half of the annual precipitation falls during the summer monsoon season (i.e., June to August). The NRE has been designated as a natural monument (No. 179), a wetland protection area, and a nature reserve due to its ecological significance as a wintering habitat for migratory waterfowl and its distinctive biodiversity.

Plant materials

Two dominant emergent plant species, *P. australis* and *B. planiculmis*, were used in the NRE pot experiments. *P. australis*, also known as common reed, is a perennial wetland plant in the Poaceae family that grows to about 1-3m high. *P. australis* is distributed in broad habitat ranges from inland areas to brackish coastal environments. In the Nakdong River Estuary, the *P. australis* community is established along the mid-high tide line of the tidal marsh, sandbars, and elevated sand dunes (Busan Metropolitan City 2022). *B. planiculmis* is a wetland plant with tuberous roots, belonging to the perennial Cyperaceae family and can grow up to a height of 0.4-0.7m. The *B. planiculmis* community is also distributed along the mid to high tide line of the tidal marsh, partly overlapping with *P. australis*. Both plant species are clonal plants, with a guerrilla architecture that provides advantages for foraging in heterogeneous environments (Xue et al. 2018). The annual production of the tuberous root of *B. planiculmis* is regionally important in NRE and the East Asian Australasian Flyway because many waterfowl species such as *Cygnus cygnus* (Whooper swan) and *Anser fabalis* (Bean goose) utilize it as a carbohydrate source during the winter season at the stopover site (Kim et al. 2013, 2016; Kim and Kim 2021).

We collected all plant material from a nearby population that was established close to the restored wetland in the NRE. To cultivate plant clones for our manipulation experiments, we first collected the rhizomes of juvenile *Phragmites* plants at the restored wetland in April 2010 (Fig. 1a). We cut the collected rhizomes to be 10cm in length and planted them in plastic pots that were 10cm in diameter and 15cm in height. We filled each pot with sediment that we collected from the tidal marsh of NRE. Collected sediments were sieved through a 2mm filter to remove other plant materials prior to use. The texture of the collected soil was classified as sandy loam. *Phragmites* rhizomes were transplanted and cultivated in soil saturated with water from the restored wetland. In October 2009, tubers of *B. planiculmis* were collected from the NRE and stored in a wet condition inside a refrigerator (4 °C). The growth rate of juvenile plants can be

impacted by different tuber sizes, so we screened tubers of the same size (i.e., length: 2 cm). In April of the following year, we planted selected tubers of *B. planiculmis* in plastic pots and kept them under saturated soil conditions for two weeks to facilitate tuber sprouting. We grew all plant clones of the wetland species under the same condition for two months before conducting the manipulation experiments.

Pot experiments

Two mesocosm tanks, measuring 6 meters in length, 4 meters in width, and 1 meter in depth, were positioned on the shore of a restored wetland in Eulsuk Island, Nakdong River Estuary (coordinates: N 35.1039°, E 128.9464°; EPSG 4326). The aforementioned restored wetland is situated near the estuarine barrage and connected to the primary river channel through two sluice gates. Therefore, the average salinity of the restored wetland remains brackish, except during prolonged flood seasons after the summer monsoon period. The mesocosm tanks were filled with water from the restored wetland. The salinity of the tank water was checked, and it was maintained at a brackish condition that ranged between 4-8 PSU during the experiment periods.

The experimental tanks were used to manipulate four different water level conditions (i.e., 0cm, 10cm, 30cm, 50cm) using bottom blocks (see Fig. 1b). The water level was set at 0 cm as a control condition. Pots of *P. australis* and *B. planiculmis* were then planted in four different water level conditions mixed with four different inundation periods (i.e., 0, 10, 20, 30 days). To compare the growth response of *P. australis* under combined stressors, we subjected *P. australis* pots, one using stem-cutting as a mechanical management option, to the restoration wetlands. There were eight replication pots for *P. australis*, including one group with stem-cutting, and 10 replication pots for *B. planiculmis* in each manipulation experiment group. We randomly placed the replicated pots within manipulated sections in the tanks.

We harvested all plants at the end of the 120-day growth period after the manipulation experiment. We measured stem length (cm), stem diameter (cm), stem density in each pot (stems per pot), and dry weight for each pot. Stem volume was calculated assuming the stem was a volumetric cone by using the following equation.

$$\text{Stem volume (cm}^3 \text{ stem} \cdot \text{pot}^{-1}) = \pi \times \text{stem radius (cm)}^2 \times \text{stem length (cm)} \div 3 \times \text{stem density (stem} \cdot \text{pot}^{-1}) \quad (1)$$

The number of pots with surviving plants was recorded to calculate the mortality ratio for different inundation depths and periods. All measured plant materials were moved to the laboratory and cleaned with tap water to remove any attached sediment or algae. The dry weight of all plant materials was measured after being dried in a 70°C convection oven for three days. In addition, the number of *B. planiculmis* tubers was counted, and their dry weight was also recorded.

Data analysis

The differences in measured response variables between *P. australis* and *B. planiculmis* were tested using the aligned-rank transform (ART) ANOVA method. The ART ANOVA is a non-parametric analysis of variance that allows for the testing of the combined treatment effects of two factors (Wobbrock et al. 2011). When the main effect was significant, post-hoc comparisons for the interaction in a two-way model were carried out using ART contrasts (ART-C; Elkin et al. 2021). All statistical analysis was performed using the R environment (version 4.3.0, R Foundation for Statistical Computing, Austria).

Results

Inundation effect on the mortality of *P. australis* and *B. planiculmis*

The mortality of two emergent plants was notably affected by different inundation depths and lengths in pot experiments (Fig. 2a-c). All plants in control groups survived under inundation treatments, with a mortality rate of 0%, except for those in the stem cutting experiment, which had a 12.5% mortality rate. Inundation treatments slightly increased the mortality rate of *P. australis* to a range of 0-37.8%, as shown in Fig. 2a. When combined with mechanical stress (i.e., stem cutting) prior to inundation treatment, these inundation effects on the mortality rate were further enhanced (Fig. 2b). The mortality rate of *P. australis* increased to 50-87.5% with both inundation and stem cutting. The mortality rate of *P. australis* was higher in groups subjected to inundation for 20-30 days when compared to those subjected to short-term inundation for 10 days. The mortality rate of *B. planiculmis* increased with deeper inundation depths and longer inundation periods (see Fig. 2c). All pots planted with *B. planiculmis* survived the 10cm inundation treatment. The mortality rate of *B. planiculmis* increased when exposed to 30-50cm inundation for 20-30 days.

Change of stem volume and biomass

We found that changes in inundation depth significantly affected the stem volume and total biomass of *P. australis* (Table 1 and Fig. 3a-b). However, the growth characteristics of *P. australis* remained unchanged with increasing inundation periods (stem volume: $F = 1.13$, $p = 0.329$; total biomass: $F = 0.33$, $p = 0.718$). When combined with stem cutting treatment, the stem volume of *P. australis* showed a significant decrease with changing inundation depths ($F = 5.99$, $p = 0.004$) and periods ($F = 6.03$, $p = 0.004$; Fig. 3c-d). After the stem cutting, all combinations of different inundation treatments showed a decrease in total biomass compared to the control group. Therefore, we did not observe any difference between the different inundation treatment groups ($p > 0.38$).

In comparison, *B. planiculmis* was found to be more sensitive to different inundation conditions than *P. australis* (Table 1 and Fig. 3e-f). The stem volume and total biomass of *B. planiculmis* decreased as the depth and duration of inundation increased. The joint effect of inundation depths and periods prompted significant changes in the growth of *B. planiculmis* across disparate inundation environments (stem volume: $F = 22.62$, $p < 0.001$; total biomass: $F = 28.04$, $p < 0.001$). In particular, the combined effect of inundation depth and period showed

distinctive growth response patterns of *B. planiculmis* (Fig. 3 e-f). At inundation depths below 10cm, the stem volume and total biomass of *B. planiculmis* increased with longer inundation periods. Under deeper inundation conditions, between 30 and 50 cm, longer inundation periods tend to reduce *B. planiculmis*' stem volume and biomass. This suggests that *B. planiculmis* thrives in shallow habitat conditions, ranging from 10 to 30 cm.

Tuber production of *B. planiculmis*

We also compared how inundation treatments affect the production of tubers in *B. planiculmis*. Similar to the results of stem volume and total biomass, the density and total weight of *B. planiculmis* tubers exhibited a negative response to shifting inundation conditions (Table 2 and Fig. 4a-b). The tuber density was higher in the 10cm inundation group and the control group than in plants that grew in deeper inundation depths ($F = 52.40$, $p < 0.001$). Tuber density increased with the inundation periods under the 10cm inundation treatment. Under deeper inundation conditions, tuber density decreased with increasing inundation periods ($F = 20.50$, $p < 0.001$). Total tuber weight exhibited a similar response pattern to the changes in tuber density under different inundation conditions.

Discussion

Effect of temporal inundation in restored wetland

We tested the growth characteristics of *P. australis* and *B. planiculmis* for the effect of temporal inundation, aboveground cutting, and the combined effect of both. Earlier studies on freshwater *P. australis* populations have reported that increasing inundation period, depth, or frequency can rapidly reduce the growth and biomass of *P. australis* (Tóth 2016; Yi et al. 2020). In this study, we observed that the independent inundation treatment, involving temporal inundation and aboveground cutting, does not significantly reduce the growth of *P. australis* when compared to the control group (i.e., saturated surface sediment). The inconsistent effects of inundation treatment observed in this study may be attributable to the local environmental conditions to which the experimental *Phragmites* species has been adapted. Song et al. (Song et al. 2021) identified the distinct local adaptations of *P. australis* populations from inland and coastal wetlands. Due to periodic environmental variations associated with the hydrological regime (such as mean water level, fluctuation frequency, and tidal range), the growth characteristics of *P. australis* populations in coastal wetlands may experience reduced physiological stress via water level manipulation.

Likewise, populations of *P. australis* distributed on intertidal mudflats in the Nakdong R. Estuary have adapted to periodic tidal changes. The range of water level manipulation (i.e., 0-50 cm) used in our inundation experiments may not have a sufficient effect to induce physiological changes in *P. australis* growth. This finding indicates that a greater degree of inundation or water level manipulation is necessary to manage the growth of *P. australis* in coastal areas, where local plant populations are consistently subjected to inundation, decreased soil iron oxides (Ding et al. 2021) and water level fluctuations. This can serve as an applicable

management measure for controlling the spread of *P. australis*, if the objective is only to control the expansion of *P. australis*. However, there may be possible differences in plant growth responses due to environmental settings between restored wetland sites and controlled mesocosms, so further field experiments are needed for in-depth understanding.

As described in our experimental site, the potential range of inundation treatment in restored wetlands may be constrained when the management objective deals with multiple species or management targets. We found that different growth responses to manipulated inundation stress were observed between *P. australis* and *B. planiculmis*. In our experiment, under 10-50cm of inundation stress, the growth inhibition of *B. planiculmis* was significantly greater than that of *P. australis*. This suggests that using inundation treatment for vegetation control may negatively impact other endemic wetland plants that have a narrower range of adaptations for water level fluctuations or inundation periods. *P. australis* has a higher density of aerenchyma in its stems, rhizomes, and roots compared to *B. planiculmis* (Mal and Narine 2004; Ding et al. 2021; Tshapa et al. 2021). The enhanced gas pathway of *P. australis* ensures the oxygen supply to the belowground system during flood conditions. These morphological characteristics explain the different growth responses of the two emergent plants in our experiments. The output provides empirical evidence that deeper inundation cannot efficiently manage to maintain higher biomass of *B. planiculmis*, an important food plant resources in the study site.

In our experiment, we observed that the biomass and tuber production of *B. planiculmis* tended to increase under shallow inundation of 0-20 cm and short-term inundation periods lasting 10-20 days. Similar experiments where water level was manipulated have also shown that tuber production in *B. planiculmis* was higher in shallow inundation environments (less than 5-10 cm), and decreased in deeper water conditions (Liu et al. 2016; An et al. 2018; Ding et al. 2021). In controlled plot experiments, An et al. reported that the optimum growth range for *B. planiculmis* in freshwater environments is between 11-36 cm depth (An et al. 2022). They also suggested that shallow inundation (0-10 cm) during the early growth season and deeper inundation (10-25 cm) during later growth seasons can assist in *B. planiculmis* growth and enable it to outcompete other plant species (An et al. 2022). This could be attributed to the local adaptation of *B. planiculmis* communities, which are often distributed in fluctuating water level environments (Hroudová et al. 2014).

Aboveground-cutting effect on the growth of *P. australis*

Previous restoration experiments reported that cutting and leaving the above-ground part of the *P. australis* resulted in rapid expansion of the remaining belowground plant portion within a year, and aboveground cutting had no significant effect (Greet and King 2019). A study using various mowing regimes, such as stem cutting, on *P. australis* plot experiments in Central Europe found that mowing only had a short-term effect on decreasing shoot size, even with various cutting trials (Güsewell et al. 2000). There seems to be weak practical evidence supporting the long-term effect of a single mowing event. Other studies have emphasized the

importance of nutrient availability in controlling excessive colonization of *P. australis* in wetland ecosystems (Uddin and Robinson 2018).

In our short-term experiment, we discovered that combining aboveground-cutting and inundation treatments has a significant restraining effect on the growth and survival rate of *P. australis*. The growth traits such as stem volume and biomass of *P. australis*, along with its associated survival rate, were both decreased. It is noteworthy that, despite an increased level of inundation after cutting, there was no increase in the growth inhibition effect. Similar application in Laurentian Great Lakes watershed showed that cut-to-drown management strategy for *P. australis* greatly reduced belowground biomass production and rhizome non-structural carbohydrate content (Widin et al., 2023). The aboveground cutting of *P. australis* not only impacts the total biomass of *P. australis* patches but also has a significant management effect in reducing the adaptation capacity of *P. australis* in wetland ecosystems. For instance, the dry shoots of *P. australis* which remain (usually produced the past year) play a significant role in aerating the belowground system through the Venturi effect (Björn et al. 2022).

Conclusions

Drawing upon two core findings from our manipulated experiment, we recommend a combined treatment approach of moderate inundation (e.g., 5-10 cm, for 20-30 days) and aboveground cutting to manage the excessive expansion of *P. australis* in restored brackish wetlands. Proposed measures can reduce unintended harm to other native wetland plant species, including *B. planiculmis*, which is a food source for migratory waterfowl, by controlling the growth of *P. australis* using moderate inundation management techniques. A low range of water level adjustment offers management benefits that minimize the compositional change of the hydrological regime, which can also affect the zonation of wetland species and their community composition. In cases where the *P. australis* patch is substantially large, the area that can be reasonably managed may be restricted due to the high resource requirement for physically removing *P. australis*. Since each restoration wetland is unique and requires tailored management approaches, further advancements in the management of large-scale *P. australis* beds are necessary.

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- 451

Table 1 (on next page)

Aligned-rank transform ANOVA results on the effects of inundation depth and periods on growth characteristics of *Pragmites australis* and *Bolboschoenus planiculmis*.

P. australis has two different treatment groups (*P. australis*: control, *P. australis* (cut): stem cutting). *: significant at p-value <0.05, **: significant at p-value <0.01, ***: significant at p-value <0.001.

		Species						
Traits	Factors	<i>P. australis</i>		<i>P. australis</i>		<i>B. planiculmis</i>		
				(cut)				
		F	p	F	P	F	p	
Stem	depth	6.74	0.002**	5.99	0.004**	49.95	<0.001***	
volume	period	1.13	0.329	6.03	0.004**	18.01	<0.001***	
	depth	x	0.95	0.443	1.05	0.387	22.62	<0.001***
	period							
Total	depth	8.38	<0.001***	0.98	0.380	66.27	<0.001***	
biomass	period	0.33	0.718	0.45	0.640	0.87	0.423	
	depth	x	1.01	0.409	2.14	0.086	28.04	<0.001***
	period							

1

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Table 2 (on next page)

Aligned-rank transform ANOVA results on the effects of inundation depth and periods on tuber production of *Bolboschoenus planiculmis*.

*: significant at p-value <0.05, **: significant at p-value <0.01, ***: significant at p-value <0.001.

Traits	Factors	<i>F</i>	p
Tuber number	depth	52.40	<0.001***
	period	3.88	0.025*
	depth x period	20.50	<0.001***
Tuber weight	depth	49.23	<0.001***
	period	0.97	0.383
	depth x period	21.64	<0.001***

1

2

Figure 1

Experimental design to test the inundation effects on the growth of two emergent species, *Pragmites australis* and *Bolboschoenus planiculmis*.

(a) experimental design, (b) illustrated experimental mesocosm established in the restored wetland.

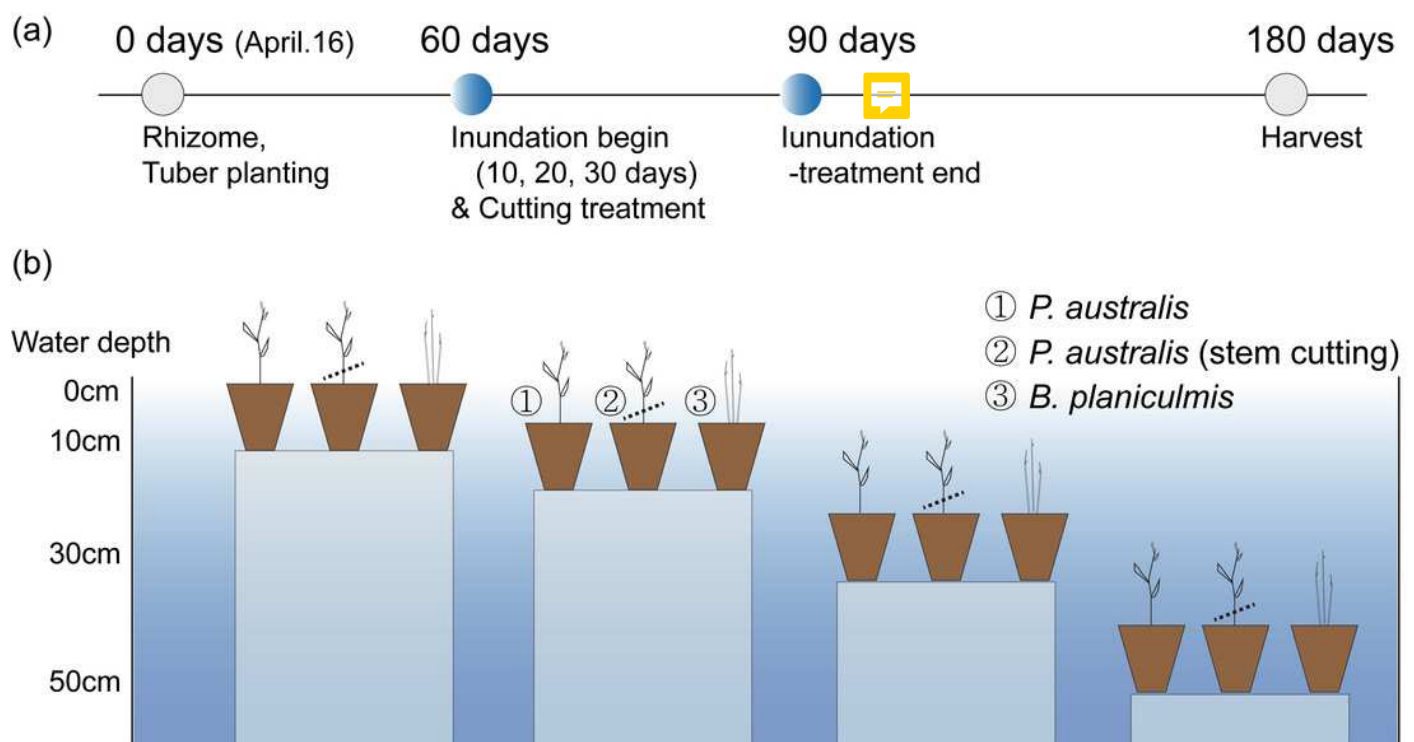


Figure 2

Mortality rate of different inundation conditions.

(a) *Phragmites australis*, (b) *P. australis* after stem cutting treatment, (c) *Bolboschoenus planiculmis*. Horizontal dot line indicates the mean of control plot (i.e., no inundation during the experimental period) of each species.

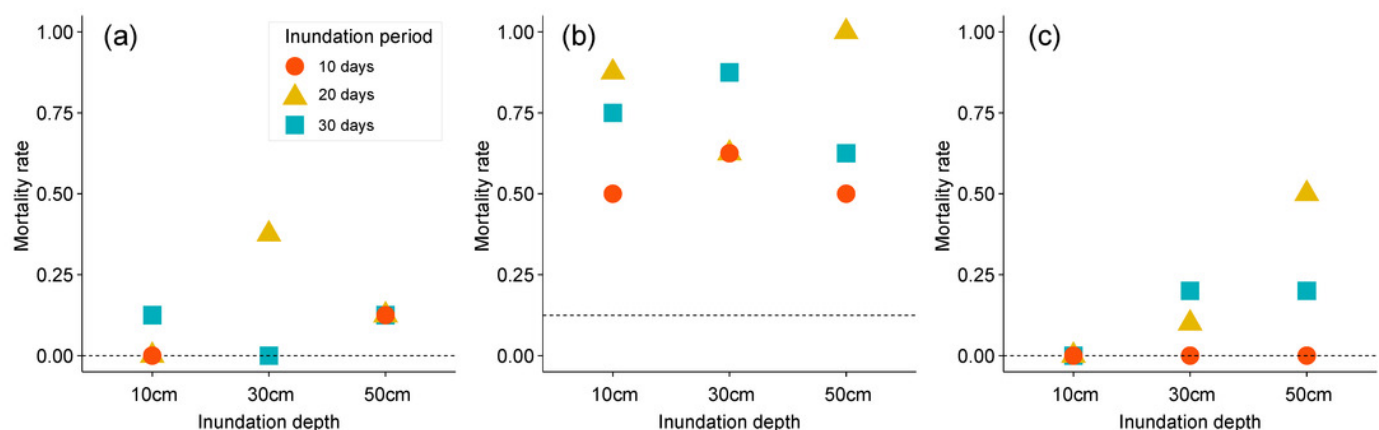


Figure 3

Stem volume and total dry weight after different inundation treatments.

(a-b) *Phragmites australis*, (c-d) *P. australis* after stem cutting treatment, (e-f) *Bolboschoenus planiculmis*. Horizontal dotted line indicates mean of the control plot (i.e., no inundation during the experimental period) of each species. NA: not available.

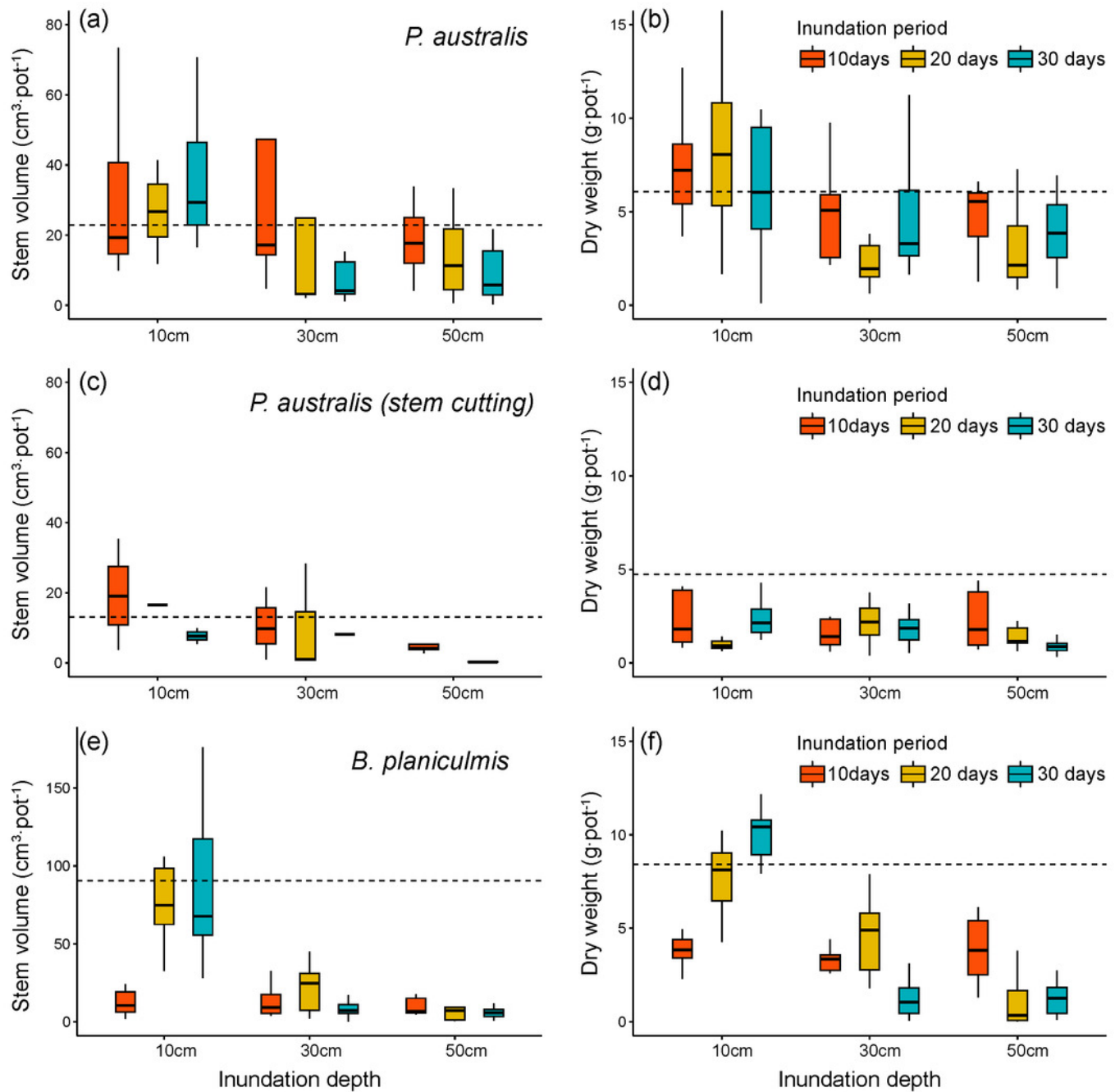


Figure 4

Tuber production of *Bolboschoenus planiculmis* under different inundation conditions.

(a) tuber density, (b) tuber dry weight. Horizontal dotted line indicates mean of the control plot (i.e., no inundation during the experimental period).

