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**Occurrence patterns of facilitation by shade along a water
gradient are mediated by species traits**

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Abstract

In disturbed habitats, shade often has facilitative effects on plants by ameliorating water and thermal stresses. Facilitation by shade tends to increase as water availability decreases. At the same time, several studies have suggested that facilitation by shade is not affected by water status or collapses under extremely dry conditions. We hypothesized that traits of beneficiary plants, specifically, the flexibility in the allocation of biomass between shoots and roots, would mediate variation in the relationship between facilitation by shade and water status. To test this hypothesis, we examined the responses of two bog species to shade under various water conditions in a post-mined peatland. The seeds of *Rhynchospora alba* and *Moliniopsis japonica* were sown under three water levels (dry: 53% peat water content, wet: 77%, and control: 71%) $\times$ two shading levels (50% shaded and unshaded). The survival, biomass, and biomass allocation between the shoots and roots of the two species were monitored for two years. Shade increased the survival and biomass of both species. However, the facilitation of *R. alba* by shade was independent of water level, whereas the strength of the facilitative effects on *M. japonica* increased as water content decreased. *R. alba* preferentially allocated biomass to roots under dry conditions and was highly drought tolerant. *M. japonica* did not alter the allocation of its biomass in response to either shade or water level and was drought intolerant. Our results suggest that flexibility in biomass allocation of beneficiary plants mediates occurrence patterns of facilitation by shade along a water gradient. The facilitation of species with inflexible biomass allocation by shade through the amelioration of water stress increases as water availability decreases, whereas the facilitation of species with flexible biomass allocation is independent of water status. Such species-specific facilitation would promote the coexistence of diverse species in a community.

Keywords: Biomass allocation; Drought; Facilitation; Shade; Post-mined peatland.

1. Introduction

Shade provided by plant canopies regulates understory plant performance and therefore plays a pivotal role in structuring plant communities (Hautier et al., 2009). In addition to increasing the strength of competition through a reduction in light availability, canopy shade facilitates understory plants by ameliorating their water and thermal stresses (Callaway, 1995; Fulbright et al., 1995). Facilitation by shade is particularly important for the establishment of plants in arid, alpine, and disturbed ecosystems characterized by frequent droughts and high ground surface temperatures, and considerable research has been conducted to understand occurrence patterns and their underlying mechanisms (Tewksbury and Lloyd, 2001; Baumeister and Callaway, 2006; Armas et al., 2011). In theory, facilitation by shade becomes stronger at higher levels of drought because the importance of ameliorating water and thermal stresses increases as water availability decreases (Holmgren et al., 1997). Many empirical studies have provided evidence that supports the predictions of this theory (Tewksbury and Lloyd, 2001; Holzapfel et al., 2006; Kikvidze et al., 2006; Seifan et al., 2010). However, contrasting results have also been reported, where facilitation by shade has been found to occur independently of water conditions (Sack and Grubb, 2002; Hastwell and Facelli, 2003; Sack, 2004) or to collapse under extremely dry conditions (Kitzberger et al., 2000; Valladares and Pearcy, 2002; Cavieres and Badano, 2009). Several factors are thought to explain the diverse relationship between facilitation by shade and water condition: the indicators used to evaluate plant performance, life-history stages, and the magnitude of shading (Holmgren and Scheffer, 2010; Martínez-Tillería et al., 2012). Nevertheless, the mechanisms underlying variation in the relationship between facilitation by shade and water status are not yet fully understood.

Despite its potential importance in modifying the occurrence patterns of facilitation by shade along a water gradient, species-specific adaptation of beneficiary plants to

environmental variation has been overlooked in previous studies. Plants respond to environmental variation in various ways; one common way is for plants to alter the allocation of their biomass between shoots and roots (Sultan, 2000). Under water-limited conditions, increasing allocation to root mass is considered to be adaptive because additional root mass improves water capture (Wilson, 1988; Bazzaz, 1996). Species that preferentially allocate biomass to roots under drought are less affected by a decrease in water, and thus, are likely to be tolerant of drought (Bazzaz, 1996). Drought-tolerant species with the ability to flexibly alter biomass allocation require amelioration of water stress by shade less than drought-intolerant species that do not have the ability to alter their biomass allocation. Therefore, we predict that occurrence patterns of facilitation by shade along a water gradient will differ between allocation-flexible and allocation-inflexible species.

In this study, we investigated the survival and biomass gain of two bog species that differ in the degree to which they can alter their biomass allocation under experimental shade with various water content levels in a post-mined peatland. Peat mining is one of the most destructive human disturbances in boreal wetlands, from which *Sphagnum* peat is excavated, along with vegetation and litter (Tuittila et al., 2000; Lavoie et al., 2005). While undisturbed *Sphagnum* peatlands do not desiccate, plant establishment in post-mined bare ground is restricted because of exposure to frequent droughts and high temperatures caused by direct sunlight (Campbell et al., 2002; Koyama and Tsuyuzaki, 2010). Therefore, water-related facilitation by canopy shade would play an important role in plant establishment in post-mined peatlands. At our study site, after *Rhynchospora alba* (L.) Vahl (Cyperaceae, the earliest colonizer of bare ground) establishes, the survival of seedlings of con- and hetero-specifics, including *Moliniopsis japonica* (Hack.) Hayata (Poaceae, later dominant), becomes much higher than those grown under bare ground conditions,

suggesting that the shade of *R. alba* canopy facilitates their seedling establishment (Egawa and Tsuyuzaki, 2011). Because *R. alba* is the earliest colonizer and expected to be better adapted to high irradiance and water stress than *M. japonica*, *R. alba* is expected to allocate its biomass more effectively under drought than *M. japonica*. We hypothesized that species exhibiting effective biomass allocation under dry conditions by investing more in roots than in shoots (*R. alba*) would be facilitated by shade independently of water conditions, whereas species with less flexible allocation traits (*M. japonica*) would be facilitated by interactions with water status through the amelioration of water stress by shade, and consequently, facilitation would increase when water availability decreased.

2. Materials and methods

2.1. Study site

The study site, Sarobetsu Mire, is located in northern Hokkaido, Japan (45°06' N, 141°42' E, 7 m a.s.l.). The mire originally stretched 27 km from North to South and 5–8 km from West to East, with an area of approximately 16,000 ha (Fujita et al., 2009). Between 1970 and 2003, *Sphagnum* peat was annually mined from the mire down to a depth of more than 3 m, over an area of 2–22 ha, using a large suction-type peat rig (Nishimura et al., 2009). The mining was conducted in a total area of more than 235 ha, and the former vegetation, litter, and seed bank in the area were completely removed along with the peat. After the extraction of high quality peat, residues were returned to the original locations. The post-mined surface was covered with open water soon after mining and subsequently the peat residue floated up and formed bare ground. Similar to other mined peatlands in northern Europe and Canada, the post-mined bare ground at our study site is characterized by harsh environments including frequent droughts, high ground surface temperatures, and high winds, restricting plant establishment (Koyama and Tsuyuzaki, 2010). However, over

the course of more than 20–40 years, the bare ground is gradually replaced by *R. alba* grassland, and eventually *M. japonica* grassland, usually within 10–20 years (Nishimura et al., 2009).

The present study was conducted in 2010 and 2011. The mean annual temperature was 6.8 °C (minimum = –6.0 °C in February, and maximum = 21.9 °C in August) in 2010, and 5.6 °C (minimum = –7.6 °C in January, maximum = 20.8 °C in August) in 2011 (Japan Meteorological Agency, 2011). The snow-free period is usually from mid-April to mid-November. Total precipitation during the growing season from May to August was 492 mm in 2010 and 275 mm in 2011. Strong winds blow at the study site; the maximum wind velocity during the snow-free period averaged 13.2 m/s with a peak gust of 27.1 m/s in 2010 and 12.4 m/s with a peak gust of 25.0 m/s in 2011. The seedlings of most species emerge during late May and late June (Egawa and Tsuyuzaki, 2011), and the seeds are dispersed in autumn until early November (Egawa et al., 2009).

2.2. Seed sowing experiment

2.2.1. Species and seed sources

Two perennial herbaceous species, *R. alba* and *M. japonica* were used for the seed sowing experiment. *R. alba* has narrow, tubal leaves and stems and develops turf. *M. japonica* has flat, long, tapering leaves and robust stems and forms dense tussocks. The height of mature plants averaged 24 cm and 45 cm for *R. alba* and *M. japonica*, respectively (Egawa, unpublished data). The seeds of the two species were collected from more than 50 individuals at the study site in autumn 2009 and were kept in paper bags at room temperature until use. Before sowing, the seeds were cold stratified for one month at 2 °C in the dark. The seed germinability of each species was assessed with 10 replicates of 50 seeds placed on moistened filter paper (Whatman #1) in Petri dishes (90 mm in diameter) in an

incubator at 5 °C /25 °C (12 h/12 h) under 12-h of white fluorescent light (23 $\mu\text{mol}/\text{m}^2/\text{sec}$) until no germination was recorded for more than two weeks. The seed germination rates averaged 63.2% for *R. alba* and 70.8% for *M. japonica*, demonstrating that the seeds were adequate for use in the seed sowing experiment.

2.2.2 Experimental design

On 18 May 2010, we established ninety-six 50 × 50 cm plots at 1-m intervals in a 16 × 6-lattice rectangular frame on bare ground mined in 1972. Sixteen plots were randomly assigned to each of six treatments in a factorial (three water levels) × (two shading levels) design. Three water levels were created by manipulating the ground surface level: mounds of up to 5-cm height, pits down to 5-cm depth, and unmanipulated controls. The ground surface layer to a depth of 5 cm at all water levels was disturbed equally to ensure the texture was uniform. Because the groundwater level averaged 10 ± 5 cm below the unmanipulated surface during June and August (Nishimura et al., 2009), the mounds were never saturated with ground water, but the pits were sometimes inundated. The manipulation of water level sometimes alters microbial community structure and nutrient conditions in peat surface layers in the long term (Jaatinen et al., 2007). However, the period of our experiment was relatively short (two years) and the effects of the manipulation of water level on peat nutrient conditions were likely to be small. Hereafter, mounds and pits are termed dry and wet treatments, respectively. Two shading levels, shaded and unshaded, were created using black sheer nets. Because sheer nets allow shading levels to be manipulated accurately and to separate below-ground competition that can mask aboveground effects of shade, the nets are widely introduced in experiments exploring the mechanisms of facilitation by canopy shade (e.g., Seifan et al., 2010; Semchenko et al., 2012). The rate of light transmission by the sheer nets was 50% of solar

radiation, which is equivalent to the shading rate of the *R. alba* canopy (Egawa and Tsuyuzaki, 2011). Structures with their tops and sides covered by sheer nets (60 × 60 × 30 cm) were fixed at four corners with bamboo stakes and plastic strings at each shading plot. The bottom of the sheer nets was placed 5 cm from the peat surface to allow wind and water transfer. The plants did not touch the top or sides of the nets during the study period. The sheer nets were expected to reduce the ground surface temperature by shading. In addition, although our shading structures allowed the free movement of wind, the plants in the nets most likely experienced reduced wind. Therefore, the shade treatment changed not only light intensity but also several other aboveground environments in a way similar to natural plant canopies, which are also known to reduce ground surface temperature and wind speed.

To assess how the topographical manipulation altered water availability, volumetric water content in the peat was monitored at a 1-cm depth for each water level at 1-h intervals from 27 June to 23 September in 2010 using moisture sensors with 5-cm probes (ECH₂O EC-5, Decagon Devices Inc., Pullman, Washington, USA). Light intensity and temperature at the peat surface were measured in three shaded and unshaded plots, with no topographical manipulation, at 1-h intervals from 4 June to 23 September 2010, by using light/temperature data loggers (HOBO pendant logger MAN-UA-002-64, Onset Computer, Bourne, Massachusetts, USA). The light data (lux) obtained were calibrated using photosynthetically active radiation (PAR, $\mu\text{mol}/\text{m}^2/\text{sec}$).

2.2.3. Seed sowing and sampling of plants

We preliminary investigated seedling density under natural *R. alba* canopies at ten randomly established 10 × 10 cm plots. Seedling density was 6.7 ± 0.6 (mean $\pm$ SE) per 100 cm² for *R. alba*, and 2.9 ± 0.4 for *M. japonica*. Based on this natural seedling density, we

set the number of seeds sown at eight per 100 cm² for *R. alba* and five for *M. japonica*. This approach allowed us to increase samples size while keeping the low natural density to minimize the risk of intraspecific competition. We sowed the seeds of both species in monoculture to avoid interspecific competition that could affect the outcomes of the experiment, although below-ground competition between *R. alba* and *M. japonica* seedlings is weak due to differences in the root depth of seedlings (Egawa and Tsuyuzaki, 2011). Accordingly, the seeds of each species were sown under a factorial (three water levels) × (two shading levels) design with eight replicates.

We used a 40 × 40 cm area in each plot for seed sowing, and 5 cm of the edges of the 50 × 50 cm plots were not used to avoid edge effects. We divided the 40 × 40 cm area in each plot into sixteen 10 × 10 cm cells. On May 31, 2010, either eight *R. alba* or five *M. japonica* seeds were sown at 3-cm intervals in each cell. The sown seeds were covered by white sheer nets (10 × 10 cm) for the first two weeks to prevent scattering and contamination of naturally dispersed seeds. The number of plants in each plot was counted at 1-week intervals from June to September 2010 and at 1-month intervals from May to August 2011. We randomly selected eight out of 16 cells in each plot, and excavated all plants in the selected cells in mid-August 2010 (first summer). Plants in the remaining eight cells in each plot were retained intact until the excavation during the same season in 2011 (second summer). Plants excavated in the first summer and the second summer were termed first-year plants and second-year plants, respectively. The excavated plant samples were kept in plastic bags and immediately transported to our laboratory. The plants were separated into shoots, roots, and reproductive organs. Each sample was dried at 70 °C for 48 hours in an oven and weighed.

2.3. Statistical analyses

To explore differences in daily mean peat water content among the three water levels, a non-parametric Friedman's repeated measures ANOVA followed by a Scheffe post-hoc test was conducted because the variance was not homogenous. We used repeated measures ANOVA to test differences in daily mean PAR and peat surface temperature between shaded and unshaded plots. Differences in germination rates among treatments were tested using a non-parametric Kruskal-Wallis test followed by a Steel-Dwass post-hoc test.

We investigated changes in plant survival, total biomass, and biomass allocation between shoots and roots (termed shoot to root [shoot:root] ratio) in response to shade and water treatments using generalized linear mixed models (GLMM). We established five competing hypotheses assuming differential effects of shade and water on plants; plant performances (survival, total biomass, and shoot:root ratio) were affected (1) by both shade and water treatments, and the effects of shade varied depending on water level, (2) by both shade and water treatments, but their effects were independent of each other, (3) by only shade, (4) by only water, and (5) neither by shade nor water treatment. Our objective in GLMM analyses was to explore which hypothesis was best supported by observations. For plant survival models, we used a binomial error distribution with a logit-link function. The response variable was the plant survival rate through to the second summer in a cell. We only used survival data of plants on the cells that remained undisturbed until the second summer. For the total biomass models, we applied a gamma distribution with a log-link function. The response variable was total biomass of a plant, which is the summed biomass including all shoots, roots, and reproductive organs. The biomass analysis was conducted separately for the first-year and second-year plants of each species. For the shoot:root ratio models, a Gaussian distribution with an identity-link function was applied. The response variable was shoot:root ratio calculated by dividing dry shoot mass by dry root mass. As well as the biomass analysis, the shoot:root ratio analysis was separately conducted for the

first-year and second-year plants of each species. In all GLMM analyses, full models included the following fixed factors: shade with two levels (shaded and unshaded), water with three levels (wet, dry, and control), and the number of conspecific plants in the cell (termed plant density). Plant density was included to reduce any confounding effects of intraspecific interactions on the outcomes of the experiment, although the low density we set would have already minimized the intraspecific competition. The number of plants germinating in the cell was used as plant density for survival, the first-year biomass, and the first-year shoot:root ratio models, and the number of overwintering individuals was used for the second-year biomass and shoot:root ratio models. Interaction terms between each pairs of fixed factors were also included in the full models. Plots were included as a random factor. We conducted stepwise model selection based on Akaike's Information Criterion (AIC) to detect the best models. Model selection approaches are particularly useful when several competing hypotheses are simultaneously confronted with data (Johnson and Omland, 2004). This approach allowed us to identify the best supported hypothesis regarding the effects of shade and water treatments on plant performance. All analyses were performed using the R statistical package (version 2.12.1, R Development Core Team, 2010).

3. Results

3.1. Environmental conditions for each treatment

The topographical manipulation significantly altered peat water content (Friedman's repeated measures ANOVA, $\chi^2_2 = 163.1$, $P < 0.001$). Daily mean water content during June and September 2010 was $52.5 \pm 0.9\%$ (mean $\pm$ SE) on the mounds (dry) and $76.6 \pm 0.3\%$ in the pits (wet), both of which were significantly different from the control ($70.4 \pm 0.7\%$) (Scheffe post-hoc test, $P < 0.001$). The water content on the mounds was low and highly

variable depending on precipitation, whereas it was high and stable in the pits (Fig. 1). The peat surface on the mounds was sometimes cracked because of severe drought, whereas the surface of the pits was sometimes inundated. Shading significantly reduced the PAR on the peat surface (repeated measures ANOVA, $F_{1, 111} = 464.7$, $P < 0.001$; Fig. 2). The shading treatment also significantly reduced peat surface temperature (repeated measures ANOVA, $F_{1, 111} = 275.0$, $P < 0.001$) which averaged 1.4 °C lower in the shaded plots than in the unshaded plots (Fig. 2).

3.2. Plant density and survival

Seed germination for both *R. alba* and *M. japonica* began two weeks after sowing and was completed within one month. The germination rates significantly differed among treatments for both species (Kruskal-Wallis test, $\chi^2_5 = 118.6$ and $P < 0.001$ for *R. alba* and $\chi^2_5 = 17.3$ and $P = 0.004$ for *M. japonica*). For both *R. alba* and *M. japonica*, the germination rate tended to increase as water content increased (Table 1), indicating that initial plant density was higher with greater levels of water content.

Two species showed different survival responses to the shade and water treatments (Table 1). The GLMM showed that the survival of *R. alba* was enhanced by shade, but not affected by water level (Table 2). This indicated that facilitation of *R. alba* survival by shade occurred independently of water level. The GLMM also revealed that plant density positively affected *R. alba* survival. In contrast to *R. alba*, shade facilitated the survival of *M. japonica* by interacting with water level, and the facilitative effect of shade increased as the water content decreased (Table 2). In the dry treatment, the survival rate through to the second summer of *M. japonica* was less than 4% in unshaded plots and improved to 23.4% in shaded plots (Table 1). Plant density did not affect *M. japonica* survival (Table 2), indicating that the reduced survival under dry × unshaded condition resulted directly from

drought stress rather than from intraspecific competition for water.

3.3. Total biomass

The differences in total biomass among treatments were larger in *M. japonica* than in *R. alba* in both the first and second years (Fig. 3). The biomass of first-year *R. alba* individuals was affected by water level and plant density, while the effects of shade were not detected (Table 2). Plant density positively affected the biomass of first-year *R. alba*, and the positive effect of plant density increased as water content decreased. The biomass of second-year *R. alba* individuals was increased by shade, water content, and plant density (Table 2). The effects of shade and water on second-year *R. alba* were independent of each other. The biomass of *R. alba* increased from the first year to the second year under all treatments, although the 2-year biomass increment was greatest under wet conditions and smallest under dry conditions (Fig. 3). More than 20 second-year *R. alba* individuals reached the reproductive stage in the control and wet plots, and two individuals reached this stage in dry plots. The biomass of first- and second-year *M. japonica* individuals was determined by shade, water level, and their interaction. In both years, shade increased the biomass of *M. japonica* more in dry conditions than in wet and control conditions (Table 2). Plant density positively affected the biomass of *M. japonica* in both the first and second years (Table 2), indicating that intraspecific interactions were facilitative throughout the study period. The biomass of *M. japonica* individuals decreased from the first year to the second year in the control and dry plots, particularly when shade was not provided (Fig. 3). No *M. japonica* individuals reproduced over the two years of growth.

No damage by herbivores was observed to leaves and stems throughout the growing season for both species, indicating that the effect of herbivores on plant biomass was negligible.

3.4. Shoot:root ratio

Contrary to total biomass, differences in shoot:root ratio among treatments were larger in *R. alba* than in *M. japonica*, particularly during the first year (Fig. 4). In the first year, *R. alba* altered the shoot:root ratio depending on the degree of shade, water level, and their interaction, as well as in response to plant density (Table 2). Shoot:root ratio of the first-year *R. alba* decreased with a decrease in water content, suggesting that this species invested more in roots than in shoots during drought. In addition, first-year *R. alba* increased investments to shoots under shade, and shoot:root ratio was highest under wet $\times$ shaded condition (Fig. 4). During the second year, biomass allocation of *R. alba* was determined only by the water level. During both years, *R. alba* individuals invested more in roots than shoots when the water content decreased. *M. japonica* did not change the shoot:root ratio in response to the degree of shade or water level during both the first and second years (Table 2). This finding suggests that *M. japonica* is not able to flexibly alter its biomass allocation between shoots and roots.

4. Discussion

The results of our experiment clearly demonstrated that the occurrence patterns of facilitation by shade along a water gradient differed between species with different abilities to flexibly alter their biomass allocation. Facilitation of *R. alba* that allocated more to root mass as water availability decreased was independent of water status, while *M. japonica* did not alter biomass allocation in response to drought and was facilitated by the interaction of shade and water level. Semchenko et al. (2012) also reported that the occurrence patterns of facilitation by shade differed between species with different biomass allocation traits. The authors found that the growth of species that allocated more to roots in full compared with

50% daylight were least facilitated by shade in temperate grasslands. Although flexibility in biomass allocation of beneficiary plants has not previously been recognized as a factor dictating the occurrence patterns of facilitation by shade, such species-specific adaptations to environmental variation has certainly played an important role in modifying facilitative interactions in plant communities.

Facilitation of *R. alba* independent of water availability may be explained by two possible mechanisms. On the one hand, shade ameliorated stress in *R. alba* proportionally across water levels. This finding has been reported in several other studies (Baruch et al., 2000; Sack and Grubb, 2002; Hastwell and Facelli, 2003; Sack, 2004). In our experiment, shade reduced the ground surface temperature by 1.4 °C and may have reduced air temperature. Reduced soil and air temperatures generally depress plant evapotranspiration and contribute to the maintenance of leaf water potential (Valiente-Banuet and Ezcurra, 1991; Larcher, 2003), which can ultimately release plants from water stress. In the case of *R. alba* that can adjust to various water conditions by altering its biomass allocation to roots, the magnitude of water stress may not have differed among water levels, and the importance of reduced temperatures by shade may have been nearly the same at all water levels. On the other hand, facilitation of *R. alba* may not be related to the amelioration of water stress. Instead, the shading nets might provide protection from wind. Plant canopies often protect understory plants from wind (Smith et al., 2003; Baumeister and Callaway, 2006) because wind can physically damage plants and negatively affect their growth and physiological performance (Ennos, 1997). At our study site, strong winds with gusts of up to 27 m/s occurred during the study period (Japan Meteorological Agency, 2011). These wind conditions likely caused more damage to *R. alba* with their narrow leaves and stems than to *M. japonica* with robust shoots. Our shading nets should have reduced the impact of wind on the plants in a manner similar to what may occur in natural plant canopies. Thus,

wind-related facilitation may have occurred independently of water level. In addition to wind, plant canopies are known to protect plants from herbivores (García and Obeso, 2003; Baraza et al., 2006). However, in our experiment, the effect of herbivores on the plants was negligible for both *R. alba* and *M. japonica*, and therefore, protection from herbivores is a factor that is unlikely to have contributed to the observed patterns of facilitation.

For *M. japonica*, the strength of facilitation by shade increased as water content decreased, and this pattern was consistent with how theory predicts facilitation by shade to occur along a water gradient (Holmgren et al., 1997). This result indicates that the most important benefit provided by shade for *M. japonica* was improvement of plant water relations. As we expected, *M. japonica* was drought-intolerant; when shade was not provided, the survival rate of *M. japonica* was less than 4% in dry conditions, while in wet conditions, the survival rate was greater than 60%. The drought-intolerance of *M. japonica* is likely attributed to their poor ability to alter their biomass allocation. Because *M. japonica* did not increase root mass under dry conditions, this species likely failed to capture enough water and suffered from strong water stress. The temperatures reduced by shade should have alleviated water stress for *M. japonica*, particularly in dry plots.

While our experiment was designed to assess the effects of shade in the absence of below-ground competition, root competition between canopy and understory plants is capable of modifying occurrence patterns of facilitation by shade under natural conditions. For instance, canopy plants sometimes engage in competition for water with understory plants, particularly in dry conditions (Valladares and Pearcy, 2002; Valladares et al., 2008). The competition for water between canopy and understory plants in dry conditions can reduce the benefits of canopy shade, and as a result, may mask the effects of aboveground facilitation by shade during drought (Holmgren et al., 2012). Further studies are required to quantify how below-ground competition between canopy and understory plants affects the

occurrence patterns of facilitation by shade along a water gradient, and how species-specific adaptations of beneficiary plants modify these patterns.

In our experiment, positive interactions among understory plants were detected for both species. In stressful ecosystems, positive density dependence in juvenile plants is often observed, most likely because the risk of physical damages decreases with an increase in neighboring plants (Anderson, 2009; Fajardo and McIntire, 2011). This mechanism likely explains our results because strong winds that were capable of damaging plants occurred at our study site. However, intraspecific interactions among understory plants may become competitive beyond the timeframe of our experiment because the strength of competition generally increases with the growth of plants (Weigelt et al., 2002). Competition among understory plants may mask the facilitative effects by shade across all levels of water content.

5. Conclusion

Our results emphasize that there is a strong relationship between the occurrence patterns of facilitation by shade and the flexibility of biomass allocation of understory plants. The facilitation of drought-intolerant species that cannot flexibly alter biomass allocation by shade through the amelioration of water stress increases with a decrease in water availability, whereas the facilitation of drought-tolerant species with flexible allocation appears to be independent of water status. These species-specific facilitation patterns may play an important role in permitting the coexistence of diverse species in a community.

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535 **Table 1**

536 Germination rate and survival rate from germination to mid-August 2010 (the first summer), April 2011 (overwintering), and mid-August
 537 2011 (the second summer) under a factorial (three water levels) $\times$ (two shading levels) design. Mean values are shown with SE. Different
 538 letters on germination rate indicate significant differences at $P < 0.05$ (Steel-Dwass post-hoc test). For results of survival analysis, see Table
 539 2.

Treatments	<i>Rhynchospora alba</i>				<i>Moliniopsis japonica</i>			
	Germination (%)	Survival (%) to			Germination (%)	Survival (%) to		
		mid-Aug 2010	April 2011	mid-Aug 2011		mid-Aug 2010	April 2011	mid-Aug 2011
Wet $\times$ unshaded	66.9 $\pm$ 4.6 ^{ab}	78.6 $\pm$ 2.3	51.1 $\pm$ 3.8	45.4 $\pm$ 4.0	68.1 $\pm$ 2.8 ^{abc}	85.1 $\pm$ 2.0	62.0 $\pm$ 4.2	61.2 $\pm$ 4.2
Wet $\times$ shaded	74.8 $\pm$ 4.4 ^a	96.6 $\pm$ 0.8	65.3 $\pm$ 3.8	61.6 $\pm$ 3.8	69.7 $\pm$ 3.6 ^{ab}	90.5 $\pm$ 1.7	73.5 $\pm$ 3.6	73.0 $\pm$ 3.7
Control $\times$ unshaded	53.7 $\pm$ 4.8 ^c	91.7 $\pm$ 1.5	73.2 $\pm$ 3.7	41.0 $\pm$ 4.6	65.6 $\pm$ 3.2 ^{abcd}	84.0 $\pm$ 2.2	47.2 $\pm$ 4.0	17.6 $\pm$ 3.1
Control $\times$ shaded	61.0 $\pm$ 5.8 ^{bc}	96.5 $\pm$ 1.0	70.5 $\pm$ 4.4	57.6 $\pm$ 4.9	60.3 $\pm$ 5.9 ^{cd}	95.7 $\pm$ 1.3	54.6 $\pm$ 4.5	31.9 $\pm$ 3.7
Dry $\times$ unshaded	43.3 $\pm$ 4.6 ^d	84.0 $\pm$ 2.3	57.2 $\pm$ 4.9	38.8 $\pm$ 5.0	59.5 $\pm$ 3.2 ^d	80.3 $\pm$ 2.4	41.1 $\pm$ 4.2	3.7 $\pm$ 1.4
Dry $\times$ shaded	37.8 $\pm$ 5.9 ^d	96.1 $\pm$ 1.2	85.4 $\pm$ 3.2	69.6 $\pm$ 4.6	65.5 $\pm$ 4.1 ^{bcd}	96.5 $\pm$ 1.0	64.5 $\pm$ 3.9	23.4 $\pm$ 3.8

Table 2

Effects of shade, water level, and plant density on the survival, total biomass, and shoot:root ratio of *Rhynchospora alba* and *Moliniopsis japonica* examined using GLMM. First year and second year indicate the age of the plants. The fixed factors in the best model based on the AIC are shown. Coefficients for shade, water level, interaction between shade and water, and interactions between plant density and shade and water were calculated with respect to the base levels shown by (0). “–” indicates a factor not selected in the best models.

Fixed factors	<i>Rhynchospora alba</i>					<i>Moliniopsis japonica</i>				
	Survival	Biomass		Shoot:root ratio		Survival	Biomass		Shoot:root ratio	
		First year	Second year	First year	Second year		First Year	Second Year	First year	Second year
Intercept	-0.893	1.746	2.866	6.369	6.623	-1.766	2.038	1.947	1.745	0.814
Shade		–			–				–	–
Unshaded	(0)		(0)	(0)		(0)	(0)	(0)		
Shaded	0.986		0.127	1.867		1.024	0.110	0.080		
Water level	–								–	–
Control		(0)	(0)	(0)	(0)	(0)	(0)	(0)		
Wet		0.435	0.109	2.612	0.052	2.293	0.253	0.447		
Dry		0.016	-0.298	-1.549	-1.667	-1.497	0.005	-0.247		
Plant density	0.106	0.044	0.081	0.213	–	–	0.027	0.092	–	–
Shade × Water level	–	–	–		–				–	–
Control × Shaded				(0)		(0)	(0)	(0)		
Wet × Shaded				1.690		-0.506	-0.173	0.065		
Dry × Shaded				0.417		0.936	0.187	0.952		
Water level × Plant density	–		–		–	–	–	–	–	–
Control × Plant density		(0)		(0)						
Wet × Plant density		-0.081		-0.369						
Dry × Plant density		0.005		0.136						
Shade × Plant density	–	–	–		–	–	–	–	–	–
Unshaded × Plant density				(0)						
Shaded × Plant density				-0.209						

Legends of figures

Fig. 1. Seasonal fluctuations in (A) water content in peat exposed to three topographical levels (wet = pit, dry = mound, and control), and (B) precipitation from June to September 2010.

Fig. 2. Daily mean (A) photosynthetically active radiation (PAR; $\mu\text{mol}/\text{m}^2/\text{sec}$) and (B) peat surface temperature ($^{\circ}\text{C}$) on shaded and unshaded plots. The bottom and top of each box show the first and third quartile, respectively, and the bold lines in the middle indicate median values. Bottom and top ends of horizontal lines represent the minimum and maximum values within the normal data range from the first quartile minus $1.5 \times$ the interquartile distance (IQD) to the third quartile plus $1.5 \times$ IQD, respectively. Outliers are indicated by open circles. Asterisks represent significant differences between unshaded and shaded conditions at $P < 0.001$ (repeated measures ANOVA).

Fig. 3. Total biomass, the sum of the shoots, roots, and reproductive organs, of the first-year and second-year plants of (A) *Rhynchospora alba* and (B) *Moliniopsis japonica* grown under a factorial (three water levels) $\times$ (two shading levels) design. The parenthetical number above each column indicates the number of reproducing individuals. For an explanation of box plots, see Fig. 2.

Fig. 4. Shoot:root ratio for the first-year and second-year plants of (A) *Rhynchospora alba* and (B) *Moliniopsis japonica* grown under a factorial (three water levels) $\times$ (two shading levels) design. For an explanation of box plots, see Fig. 2.

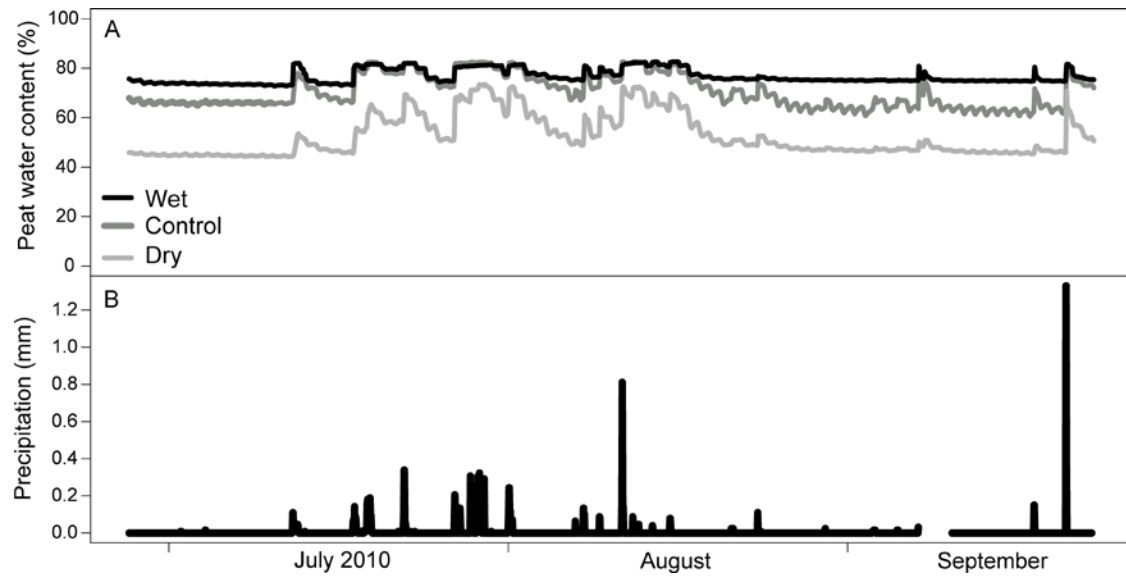


Fig. 1.

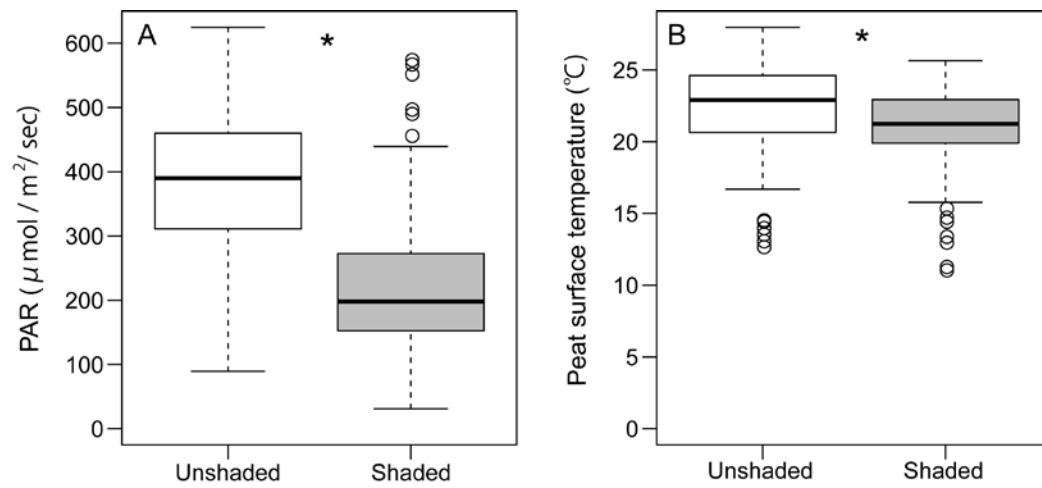


Fig. 2.

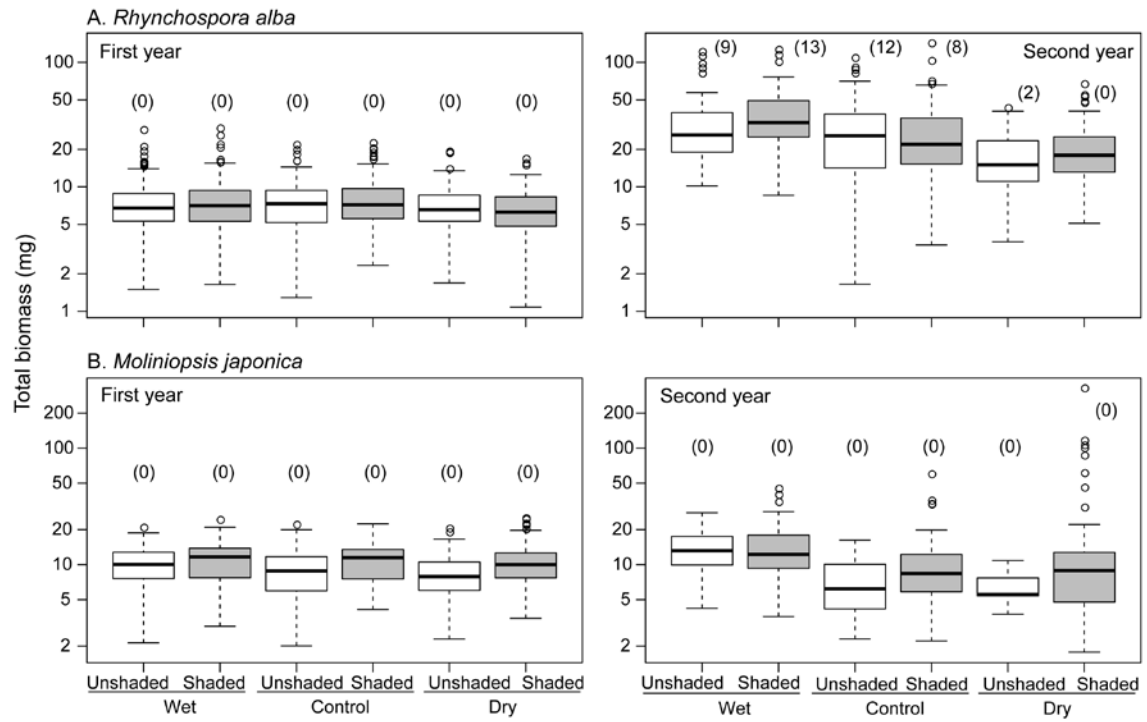


Fig. 3.

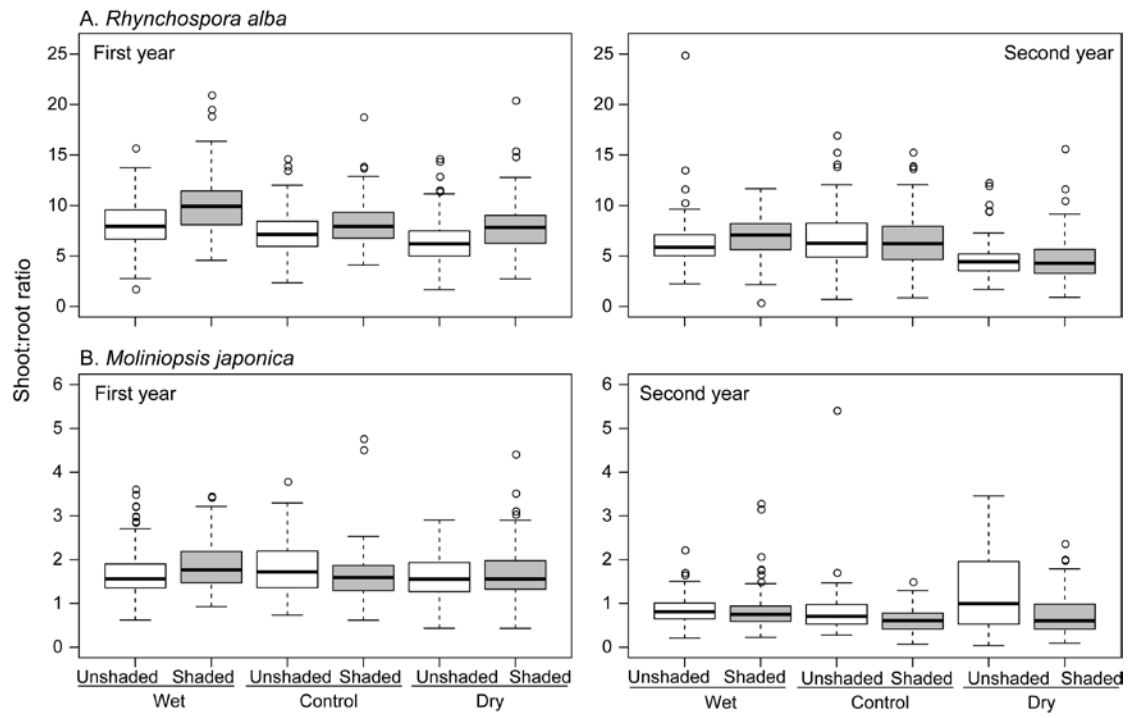


Fig. 4.