

How internode length, position and presence of leaves affect survival and growth of *Alternanthera philoxeroides* after fragmentation?

Bi-Cheng Dong · Guo-Lei Yu · Wei Guo · Ming-Xiang Zhang ·
Ming Dong · Fei-Hai Yu

Received: 1 January 2010 / Accepted: 29 April 2010 / Published online: 19 May 2010
© Springer Science+Business Media B.V. 2010

Abstract Disturbance is common in nature and disturbance-caused fragmentation of clones happens frequently in stoloniferous plants. After fragmentation storage in stolon internodes and leaves may enhance survival and growth of stoloniferous plants. We hypothesize that (1) increasing length of the internode attached to the ramet and (2) presence of leaves will increase ramet survival and growth, and that (3) internode positions (before or after the ramet or both) will also play a role. We tested these hypotheses with the stoloniferous, invasive herb *Alternanthera philoxeroides*. In one experiment, we measured survival and growth of the ramets either without stolon internode (0 cm in length) or attached with internodes of 2, 4, 6 and 8 cm and either with or without leaves. In the other experiment, we measured survival and growth of the ramets attached with a proximal internode (before the ramet), a distal internode (after the ramet) or both. Increasing internode length and presence of leaves significantly increased the survival rate and growth (biomass, leaf area, number of ramets, stolon length and number of leaves) of the *A. philoxeroides* plants. All growth measures of *A. philoxeroides* at harvest were larger when the ramets were attached with a distal internode than when they were attached with a proximal internode, but the survival rate was lower. These results support the hypotheses and suggest that storage in stolons and leaves may be of great significance for clonal plants in frequently disturbed habitats and may contribute greatly to the invasiveness of *A. philoxeroides*.

Keywords Alligator weed · Clonal growth · Clone fragmentation · Disturbance

B.-C. Dong · W. Guo · M.-X. Zhang · F.-H. Yu (✉)
College of Nature Conservation, Beijing Forestry University, P.O. Box 159,
Qinghuadonglu 35, Haidian District, Beijing 100083, China
e-mail: feihaiyu@bjfu.edu.cn

G.-L. Yu · M. Dong
State Key Laboratory of Vegetation and Environmental Change, Institute of Botany,
Chinese Academy of Sciences, Beijing 100093, China

Introduction

Disturbance is a common feature of natural habitats and occurs at various spatial and temporal scales (Klimešová and Klimeš 2003, 2008; Martínková et al. 2004; Bornette et al. 2008; Puijalón et al. 2008; Latzel and Klimešová 2009). Severe disturbance can cause destruction of above-ground plant parts (Klimešová and Klimeš 2003, 2008) and fragmentation of clones formerly interconnected by e.g. stolons, rhizomes or roots (Sainty et al. 1998; Stuefer and Huber 1999; Jia et al. 2008). In frequently disturbed habitats, therefore, plants must have developed some strategies to cope with disturbance (Klimešová and Klimeš 2003, 2008; Martínková et al. 2004; Bornette et al. 2008; Puijalón et al. 2008; Latzel and Klimešová 2009).

Stoloniferous herbs represent a large group in clonal plants and are distributed in a wide range of habitats (Klimeš et al. 1997). When remaining intact, stolon internodes play important roles in clones' foraging responses (Birch and Hutchings 1994; Wijesinghe and Hutchings 1999) and resource sharing (Alpert 1999; Alpert et al. 2003), thereby contributing greatly to their fitness (Alpert 1999; van Kleunen and Stuefer 1999; Alpert et al. 2003; Roiloa et al. 2007; Wang et al. 2008; Yu et al. 2009).

Reserves in plant organs may help plants survive and regrowth after damage caused by various disturbances such as grazing, mowing, trampling, water currents and human activities (Lawson et al. 2000; Goulas et al. 2001; Klimešová and Klimeš 2003, 2008; Martínková et al. 2004; Puijalón et al. 2008). In natural habitats stoloniferous herbs face frequently with severe disturbance that often results in fragmentation of the clones (Sainty et al. 1998; Stuefer and Huber 1999; Jia et al. 2008). After fragmentation stolon internodes may play an important role in ramet survival and growth because they may store substantial amount of non-structural carbohydrates and soluble proteins that can be mobilized and used by the ramets (Fry 1993; Bouchart et al. 1998; Stuefer and Huber 1999; Suzuki and Stuefer 1999; Lawson et al. 2000; Goulas et al. 2001). Such a role of stolon internodes may be especially important for juvenile ramets that have not rooted and thus lack the capacity to acquire water and nutrients from soils (Stuefer and Huber 1999).

Because disturbance occurs at different scales with different degrees of severity (Klimešová and Klimeš 2003, 2008; Bornette et al. 2008), stolons of clonal plants may be subjected to different degrees of damage. After fragmentation, therefore, a ramet can be attached (a) with various lengths of stolon internodes, (b) with a proximal internode (before and thus developmentally older than the ramet) only, a distal internode (after and thus developmentally younger than the ramet) only or both, and (c) with or without leaves. If the reserve in internodes contributes to the fitness of individual ramets (Stuefer and Huber 1999), then increasing length of the stolon internodes attached to the ramets will increase ramet survival and growth.

Besides the main function of photosynthesis, leaves can also store a certain amount of carbohydrates and proteins (Granstedt and Huffaker 1982; Danckwerts and Gordon 1989; Corre et al. 1996). After fragmentation, storage in leaves may also contribute to the survival and growth of unrooted ramets, especially when the storage in other plant parts (e.g. stolon internode) is insufficient. We, therefore, hypothesize that presence of leaves will contribute to ramet survival and growth and such effects will be larger when the stolon internode attached to the ramet is shorter.

Studies have shown that structural and physiological constraints can greatly affect resource mobilization within a clone (Watson and Casper 1984; Price and Hutchings 1992; Price et al. 1996; Marshall and Price 1997). For instance, in *Glechoma hederacea* vascular architecture causes sectorial patterns of resource distribution, and in some species discrete

integrated physiological units (i.e. sectorial compartments where the metabolite movement is confined; Watson and Casper 1984; Marshall and Price 1997) can be formed. Tracer studies have also shown that the efficiency of resource mobilization differs between basipetal and acropetal directions (Marshall 1990; Jonsdottir and Watson 1997). Moreover old (proximal) and young (distal) stolon internodes may differ in the function of carbohydrate storage (Turner and Pollock 1998). Thus, it is hypothesized that the positions (proximal, distal or both) of the stolon internode attached to the ramet will affect ramet survival and growth.

We tested these hypotheses in two greenhouse experiments with the stoloniferous, invasive herb *Alternanthera philoxeroides* (alligator weed). In one experiment, we measured the survival rate and growth of the ramets either without stolon internode (0 cm in length) or attached with internodes of 2, 4, 6 and 8 cm and either without or with two opposite leaves. We predict that increasing length of the internode attached to the ramet and presence of leaves will increase the survival and growth of *A. philoxeroides*. In the other experiment, we measured the survival rate and growth of the ramet attached with a proximal internode, a distal internode or both, keeping the total length of the internode segment the same. We predict that stolon internode position will significantly affect the survival and growth of *A. philoxeroides*.

Materials and methods

The species

Alternanthera philoxeroides (Mart.) Griseb. is a perennial herb of the Amaranthaceae family and originates from South America (Sainty et al. 1998; Pan et al. 2007). It is listed as one of the worst invasive weeds in many countries, including China (Holm et al. 1997; Wang et al. 2008). *A. philoxeroides* produces monopodially growing stolons, and relies mainly on clonal growth to sustain and renew its populations (Pan et al. 2007; Yu et al. 2009). A ramet of *A. philoxeroides* consists of a node that can root at moist conditions, two opposite leaves, and two axillary buds that can develop into new stolons and ramets or, at some conditions, inflorescences. *A. philoxeroides* forms white, capitate inflorescences of 1–2 cm in diameter from leaf axils, and produce viable seeds in its native range (Pan et al. 2007). However, in China it does not produce viable seeds under field conditions and genetic diversity of the populations is extremely low, likely originated from a single genotype (Ye et al. 2003; Wang et al. 2005; Pan et al. 2007). New stolons are formed throughout the growth season, whereas inflorescences are formed during summer and autumn. *A. philoxeroides* is amphibious and grows vigorously in both aquatic (e.g. still water and flowing stream) and terrestrial (e.g. roadside, cropland and lawn) environments (Sainty et al. 1998; Ma and Wang, 2005; Pan et al. 2006; Jia et al. 2008). Morphology, anatomy and growth of *A. philoxeroides* in aquatic conditions differ significantly from those in dry, terrestrial habitats, showing respective adaptations to local environments (Tao et al. 2004; Tao and Jiang 2004). But in China these differences are most likely plastic responses rather than genetically based changes (Ye et al. 2003; Wang et al. 2005; Pan et al. 2007). In aquatic situations, *A. philoxeroides* can occupy the entire water surface, restrict the growth of fish and inhibit boat movement, where it also provides a habitat for mosquitoes to breed. In terrestrial situations, *A. philoxeroides* can displace native plants, and block irrigation and drainage systems of farmland (Sainty et al. 1998; Ma and Wang 2005; Pan et al. 2006; Wang et al. 2008).

Stolon internodes of *A. philoxeroides* can be easily broken and its leaves lost due to disturbance such as herbivory, mowing, trampling, herbicides, strong water current, fishing and transportation boats, and the whole clones can be readily split into segments of different size (Sainty et al. 1998; Schooler et al. 2006; Julien et al. 2007). Because each node (ramet) has the potential to survive independently and to produce new stolons and ramets, *A. philoxeroides* can spread vigorously (Holm et al. 1997; Sainty et al. 1998).

Experimental design

Ramets of *A. philoxeroides* were collected from Jiangxi province, China in mid April, and vegetatively propagated in an artificial pond (7.0 m × 1.2 m × 1.2 m, length × width × depth) in the Botanical Garden of the Institute of Botany, the Chinese Academy of Sciences in Beijing. After about 5 months of cultivation, the surface of the pond was fully occupied by the stolons and ramets of *A. philoxeroides*. On 14 October, about 3,000 stolon segments of similar stolon diameter were cut off from this population. Each stolon segment contained an unrooted ramet (i.e. a node with two leaves and two axillary buds), the proximal internode (connected to its adjacent, older ramet) and the distal internode (connected to its adjacent, younger ramet). Both proximal and distal internodes were at least 5 cm long. Of the 3,000 stolon segments, 1,260 were selected and used for the experiments and 45 for initial measurements.

The study consisted of two experiments. The first experiment (Exp. 1, Table 1) was to test the effects of stolon internode length and presence of leaves on the survival and growth of the *A. philoxeroides* ramets. There were five treatments of stolon internode length (0, 2, 4, 6, and 8 cm), combined with two treatments of leaf presence status (with or without the two leaves). In this experiment, the stolon internode (segment) consisted of both proximal and distal internodes, each having half of the total length. For instance, for the 4 cm length treatment, both the distal and the proximal internode of the segment were cut into 2 cm long. The second experiment (Exp. 2, Table 1) was mainly to test the effects of stolon internode position and its potential interactive effects with stolon internode length and with presence of leaves. In this experiment, there were three levels of stolon internode position (the internode was located only in the proximal part—before the ramet, only in the distal part—after the ramet, or in both), combined with two levels of internode length (2 and 4 cm) and two levels of leaf presence status (with or without the two leaves). Because the four treatments in which the ramets were attached with both a proximal and a distal internode (Table 1, shaded portion) were already included in Exp. 1, we omitted them in Exp. 2. Thus, these four treatments were shared by the two experiments (Table 1, shaded portion).

In the two experiments, 63 plastic containers (39 cm × 26 cm × 10.5 cm; length × width × height) were used, and each container was divided into two equal parts, corresponding to two boxes (39 cm × 13 cm × 10.5 cm). The substrate used in the container was a mixture of sand and peat at a volume ratio of 1:1. Each container was planted with a total of 20 stolon segments: ten stolon segments with leaves were planted in one box and ten without leaves (leaves removed) were in the other. The ten segments in each box were placed horizontally in ten evenly spaced positions and then covered by 1 cm deep vermiculite, which was sufficient to completely cover all the segments and the leaves. Thus the treatments mimicked the situations faced by *A. philoxeroides* in frequently disturbed terrestrial habitats. Covering with vermiculite had three advantages: (1) it could prevent light to reach the segments and the leaves so that the confounding effects of photosynthesis by them were avoided, (2) it would not produce a significant physical

Table 1 Design of the two experiments

Internode length (cm)	Internode position					
	Proximal		Both		Distal	
	With leaves	No leaves	With leaves	No leaves	With leaves	No leaves
0			With leaves	No leaves		
2	With leaves	No leaves	With leaves	No leaves	With leaves	No leaves
4	With leaves	No leaves	With leaves	No leaves	With leaves	No leaves
6			With leaves	No leaves		
8			With leaves	No leaves		

Exp. 1

Exp. 2

Experiment 1 (Exp. 1) had a two-factorial design and test the effects of stolon internode (segment) length (0, 2, 4, 6 and 8 cm) and presence of leaves (with or without), without considering the effect of internode position; Experiment 2 (Exp. 2) had a three-factorial design and mainly test the effects of internode position (proximal, distal or both) and its potential interactive effects with internode length (2 and 4 cm) and presence of leaves. The treatments in the shaded portion were shared by the two experiments

Table 2 ANOVA results of effects of stolon internode length (0, 2, 4, 6 and 8 cm), presence of leaves and the interaction on the survival and growth of the *Alternanthera philoxeroides* plants at harvest (on week 9)

Effect	DF	Survival rate	Biomass	Leaf area
Length	4,60	25.6***	41.5***	31.9***
Leaves	1,60	7.2**	90.4***	61.2***
Interaction	4,60	1.3 ^{ns}	0.4 ^{ns}	0.2 ^{ns}

Degree of freedom (DF), *F* values and the significance levels (*** $P < 0.001$, ** $P < 0.01$, * $P < 0.05$, and ^{ns} $P \geq 0.05$) are given

barrier as was done by the real soil, and (3) it could make moisture conditions more homogeneous and comparable across the treatments. In frequently disturbed habitats, it is also possible that the ramets are covered by soils after clone fragmentation. During the experiments, the containers were systematically repositioned to avoid effects of possible environment patchiness within the greenhouse. To keep the soil moist, tap water was supplied once a day. There were seven replicates of boxes for each of the 18 treatments in the two experiments (Table 1).

The experiments started on 15 September 2009, ended 9 weeks later on 20 November, and were conducted in a heated greenhouse at Forest Science Co, Ltd of Beijing Forestry University. During the experiments mean temperature in the greenhouse were 17.4°C, as measured hourly by two Hygrochron temperature loggers (iButton DS1923; Maxim Integrated Products, USA).

At the start of the experiments, the average diameter of the stolon segments was 1.92 ± 0.24 mm (mean \pm SD, $N = 40$). The dry weight of the 0-, 2-, 4-, 6- and 8-cm-long stolon segments were 6.9 ± 2.0 , 8.4 ± 2.9 , 12.1 ± 4.1 , 15.8 ± 2.5 , 17.0 ± 2.0 mg (mean \pm SD, $N = 5$), respectively.

Measurements

On 4–5 October (week 3), 31 October–1 November (week 6) and 18–19 November (week 9), number of ramets and number of leaves were counted and total stolon length was measured for each *A. philoxeroides* plant that had emerged from the soil layer. On 20–22 November, each living plant was harvested and separated into leaves, stolons and roots. Total leaf area of each plant was measured by WinFOLIA Pro 2004a (Regent Instruments Inc., Canada). Then all plant parts were oven-dried at 70°C for 48 h and weighed. Whether the initial internode still existed or was depleted was also noted during harvest.

Data analysis

Because each box was initially planted with ten stolon segments, we could calculate the survival rate of the ramets in each box at harvest. We also calculated the average number of ramets, number of leaves, stolon length, leaf area and biomass of the emerged, living *A. philoxeroides* plants in each box. These data (survival rate and mean values of the variables measured in each box) were used in the analyses described below.

For Exp. 1, two-way ANOVA was used to investigate the effects of stolon internode length (0, 2, 4, 6 and 8 cm) and presence of leaves (with or without) on the survival rate, biomass and total leaf area of the *A. philoxeroides* plants. For Exp. 2, three-way ANOVA was conducted to test the effects of stolon internode position (proximal, distal and both), internode length (2 and 4 cm) and presence of leaves on the survival rate, biomass and leaf area of the *A. philoxeroides* plants. Data on number of ramets, stolon length and number of leaves measured on week 3, 6 and 9 were analyzed using repeated measures ANOVA, with measuring week as the repeated variable. SPSS 17.0 software (SPSS, Chicago, IL, USA) was used for all analyses.

Results

Effects of internode length and presence of leaves: Exp. 1

Stolon internode length and presence of leaves significantly affected the survival rate, biomass, total leaf area, number of ramets, total stolon length and number of ramets of the *A. philoxeroides* plants (Tables 2 and 3, significant effects of L and Le). All these variables increased markedly with increasing stolon internode length, and were significantly larger when the leaves were present (Figs. 1 and 2). There was no interaction effect of internode length by presence of leaves (Tables 2 and 3, no significant effect of L × Le or W × L × Le).

Effects of internode position: Exp. 2

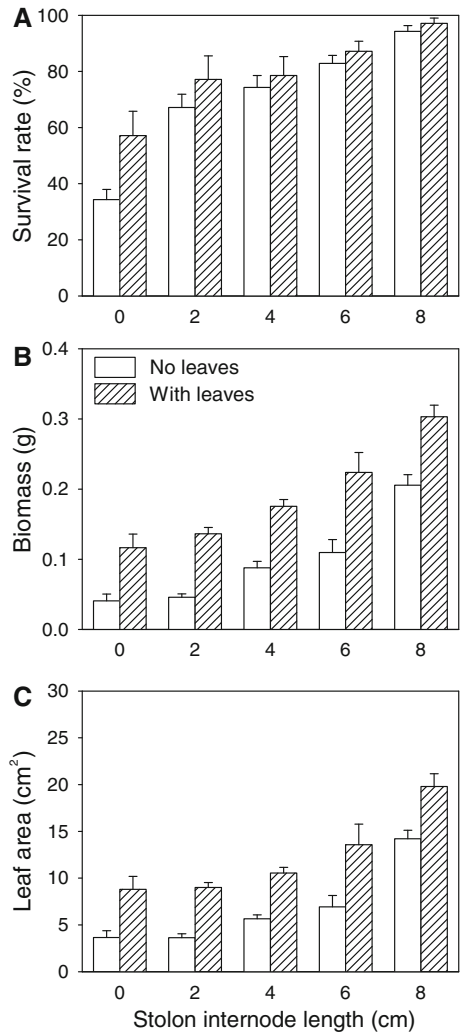
Stolon internode position significantly affected the survival rate, biomass, total leaf area, number of ramets, total stolon length and number of leaves of the *A. philoxeroides* plants (Tables 4 and 5, significant position effect). The survival rate was significantly smaller when the ramets were attached with a distal internode than when attached with a proximal internode or both (Fig. 3a). However, biomass and total leaf area were the largest when the

Table 3 ANOVA results of effects of stolon internode length (0, 2, 4, 6 and 8 cm), presence of leaves, measuring week (3, 6 and 9) and their interactions on number of ramets, stolon length and number of leaves of the *Alternanthera philoxeroides* plants

Effect	DF	No. of ramets	Stolon length	No. of leaves
Length (L)	4,60	19.6***	42.2***	17.8***
Leaf (Le)	1,60	22.9***	65.8***	36.4***
L × Le	4,60	0.2 ^{ns}	0.5 ^{ns}	0.3 ^{ns}
Measuring week (W)	2,120	612.4***	481.4***	172.5***
W × L	8,120	9.1***	14.4***	5.6***
W × Le	2,120	15.7***	20.0***	18.4***
W × L × Le	8,120	0.6 ^{ns}	1.2 ^{ns}	1.4 ^{ns}

F values and the significance levels (*** $P < 0.001$, ** $P < 0.01$, * $P < 0.05$ and ^{ns} $P \geq 0.05$) are given

Fig. 1 Effects of stolon internode length and presence of leaves on **a** survival rate, **b** biomass and **c** total leaf area of the *Alternanthera philoxeroides* plants at harvest (on week 9). Means + 1 SE are given



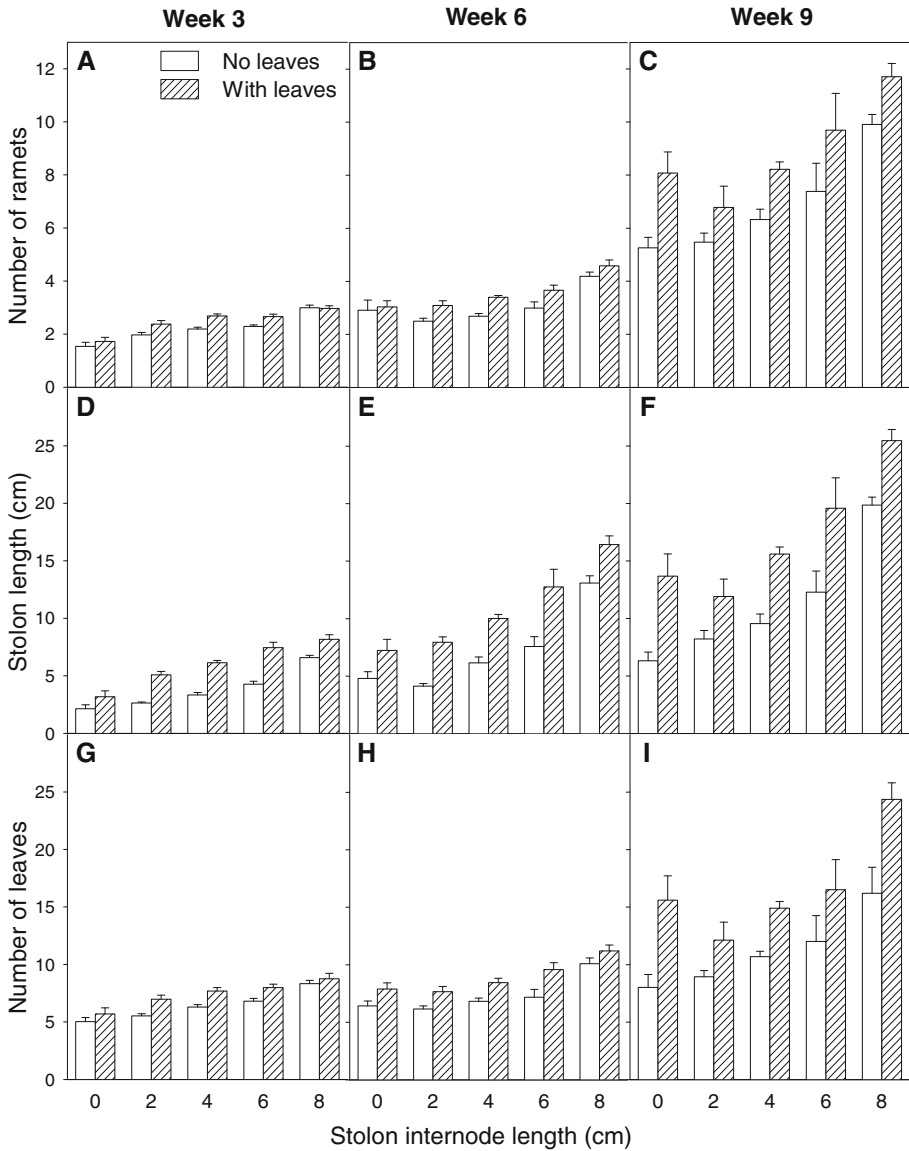


Fig. 2 Effects of stolon internode length and presence of leaves on **a–c** number of ramets, **d–f** total stolon length and **g–i** number of leaves of the *Alternanthera philoxeroides* plants measured on week 3, 6 and 9, respectively. Means + 1 SE are given

ramets were attached with a distal internode, smallest when attached with a proximal internode and intermediate when attached with both (Fig. 3b–c). On week 3, number of ramets, total stolon length and number of leaves were the largest when the ramets were attached with both internodes (Fig. 4a, d, g), but on week 6 and 9, they were the largest when the ramets were attached with a distal internode (Fig. 4b–c, e–f, h–i).

Table 4 ANOVA results of effects of stolon internode position, length (2 and 4 cm), presence of leaves and their interactions on the survival and growth of the *Alternanthera philoxeroides* plants at harvest (on week 9)

Effect	DF	Survival rate	Biomass	Leaf area
Position (P)	2,72	6.3**	36.8***	23.8***
Length (L)	1,72	9.4**	93.6***	43.1***
Leaves (Le)	1,72	24.1***	320.4***	206.3***
P × L	2,72	1.0 ^{ns}	11.5***	5.0**
P × Le	2,72	2.8 ^{ns}	9.8***	8.9***
L × Le	1,72	3.7 ^{ns}	1.4 ^{ns}	0.0 ^{ns}
P × L × Le	2,72	0.6 ^{ns}	4.5*	2.5 ^{ns}

Degree of freedom (DF), *F* values and the significance levels (*** $P < 0.001$, ** $P < 0.01$, * $P < 0.05$ and ^{ns} $P \geq 0.05$) are given

Table 5 ANOVA results of effects of stolon internode position, length (2 and 4 cm), presence of leaves, measuring week (3, 6 and 9) and their interactions on number of ramets, stolon length and number of leaves of the *Alternanthera philoxeroides* plants

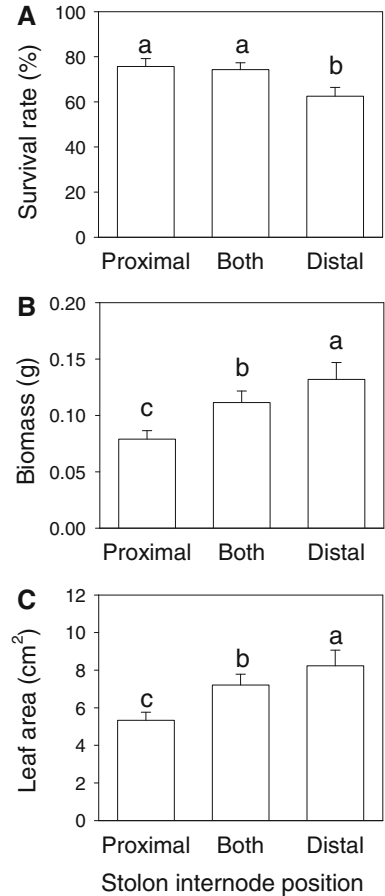
Effect	DF	No. of ramets	Stolon length	No. of leaves
Position (P)	2,72	29.4***	32.8***	37.9***
Length (L)	1,72	46.6***	111.1***	39.4***
Leaf (Le)	1,72	92.1***	293.6***	119.3***
P × L	2,72	7.2***	22.0***	7.7***
P × Le	2,72	3.5*	6.2**	3.7*
L × Le	1,72	0.4 ^{ns}	3.6 ^{ns}	0.1 ^{ns}
P × L × Le	2,72	0.2 ^{ns}	2.7 ^{ns}	0.7 ^{ns}
Measuring week (W)	2,144	1303.4***	607.0***	415.9***
W × P	4,144	31.9***	25.0***	38.1***
W × L	2,144	23.8***	15.4***	9.5***
W × Le	2,144	41.0***	30.6***	36.1***
W × P × L	4,144	8.2***	6.6***	7.3***
W × P × Le	4,144	2.1 ^{ns}	3.5**	5.4***
W × L × Le	2,144	1.2 ^{ns}	0.9 ^{ns}	0.4 ^{ns}
W × P × L × Le	4,144	0.4 ^{ns}	1.8 ^{ns}	0.9 ^{ns}

F values and the significance levels (*** $P < 0.001$, ** $P < 0.01$, * $P < 0.05$ and ^{ns} $P \geq 0.05$) are given

Interactive effects of internode position by length and by presence of leaves: Exp. 2

Effects of stolon internode position on the survival rate of the *A. philoxeroides* plants did not depend on presence of leaves or internode length (Fig. 5a, b, Table 4, no significant P × Le effect). On the other hand, the effects of internode position on biomass, total leaf area, total stolon length and number of leaves of the *A. philoxeroides* plants depended significantly on both internode length (Tables 4 and 5, significant P × L effects) and presence of leaves (Tables 4 and 5, significant P × Le effects). The effects were smaller when the stolon internode was shorter or when the leaves were removed (Fig. 5c–l).

Fig. 3 Effects of stolon internode position on **a** survival rate, **b** biomass and **c** total leaf area of the *Alternanthera philoxeroides* plants at harvest (on week 9). Bars sharing the same letters are not different at $P = 0.05$ (by Duncan's test). Means + 1 SE are given



Discussion

Few studies directly tested the roles of stolon internodes and internode positions in ramet survival and growth after severe disturbance (Stuefer and Huber 1999; Suzuki and Stuefer 1999). We found that attachment with internodes and leaves contributed greatly to ramet survival and growth of *A. philoxeroides*, and more intriguingly, the contribution by proximal and distal internodes differed significantly and also depended on internode length and presence of leaves.

The survival rate was on average 46% for the *A. philoxeroides* ramet without stolon internode and increased to 96% when it was attached with an 8-cm-long internode (Fig. 1a). Also, all growth measures increased markedly with increasing internode length (Figs. 1b, c and 2). These results agree with previous findings on *Potentilla anserina* (Stuefer and Huber 1999) and suggest that storages in stolon internodes increase the fitness of juvenile ramets. By monitoring changes in carbohydrate and protein contents in stolon internodes, it has been shown that substantial amount of non-structural carbohydrates and soluble proteins stored in stolon internodes could be readily mobilized and used for the regrowth of damaged tissues (Danckwerts and Gordon 1989; Corre et al. 1996; Baur-Höch et al. 1999; Lawson et al. 2000; Goulas et al. 2001), and that at low temperature the

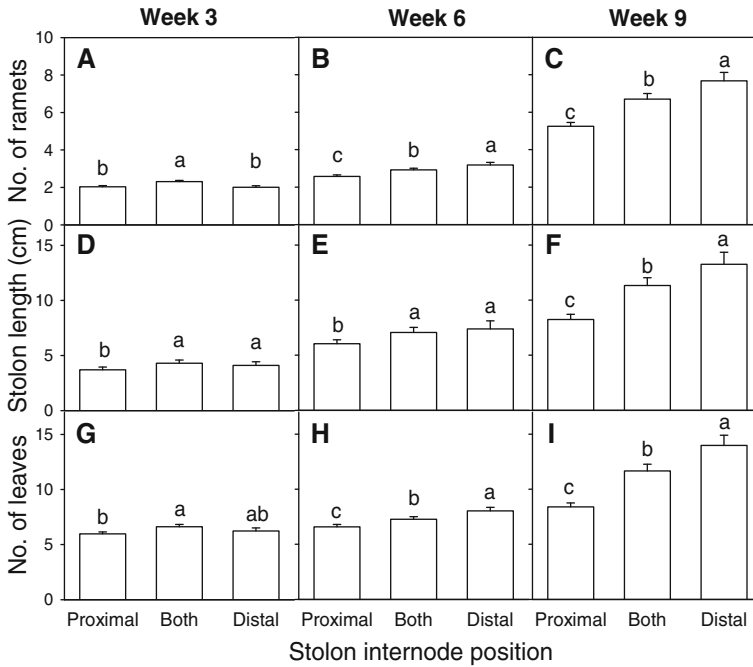


Fig. 4 Effects of stolon internode position on **a–c** number of ramets, **d–f** total stolon length and **g–i** number of leaves of the *Alternanthera philoxeroides* plants measured on week 3, 6 and 9, respectively. Bars sharing the same letters are not different at $P = 0.05$ (by Duncan’s test). Means + 1 SE are given

survival rate of the *Eremochloa ophiuroides* stolons was positively correlated with the carbohydrate content in stolon internodes (Fry 1993). Although we did not measure the mobilization of carbohydrates and nitrogen in *A. philoxeroides*, we found that 87.2% of the internodes attached to the ramets were depleted completely at the time of harvest (Dong and Yu unpubl. data). Thus, it is very likely that the carbohydrates and nitrogen stored in the stolon internodes were mobilized and used by the juvenile, unrooted ramets of *A. philoxeroides*, which greatly increased their survival and growth. In frequently disturbed habitats, therefore, efficient use of the reserves in stolon internodes may be an adaptive strategy for stoloniferous plants to cope with severe disturbance that often causes fragmentation of clones.

Presence of leaves increased the survival and growth of *A. philoxeroides*, but it did not interact with stolon internode length (Table 2, Figs. 1 and 2), suggesting that storages in leaves contribute to the fitness of juvenile ramets and such effects do not depend on the amount of the reserves in the internodes of the ramets. There is evidence that leaves can store some non-structural carbohydrates and soluble proteins (Granstedt and Huffaker 1982; Danckwerts and Gordon 1989; Corre et al. 1996), and these substances stored in the two opposite leaves may have been used by the ramets of *A. philoxeroides* to sustain its survival and growth. Therefore, in addition to the main function as a primary carbohydrate producer through photosynthesis, leaves may also serve as a short-term reserve that can be used by the ramets bearing them to deal with severe disturbance.

At harvest all growth measures of *A. philoxeroides* were larger when the ramets were attached with a distal internode than when they were attached with a proximal internode

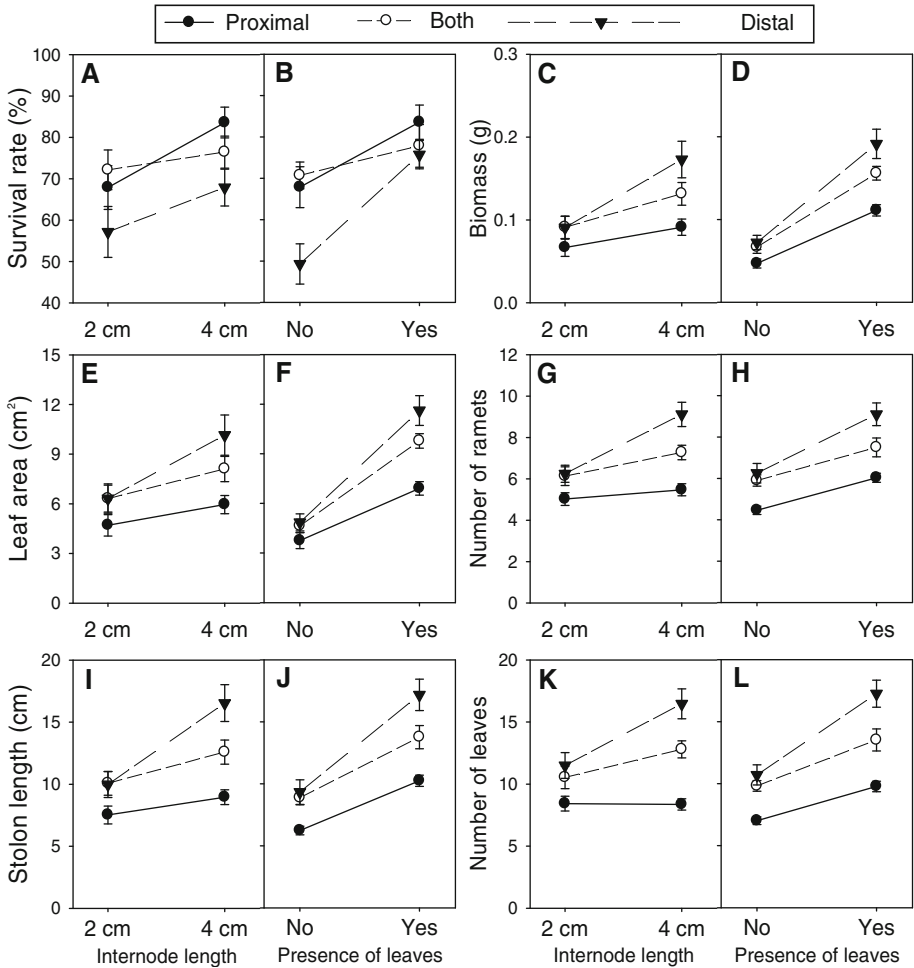


Fig. 5 Interactive effects of stolon internode position by length (a, c, e, g, i and k) and position by presence of leaves (b, d, f, h, j and l) on the survival (a–b) and growth (c–l) of the *Alternanthera philoxeroides* plants at harvest (on week 9). Means + 1 SE are given

(Figs. 3b, c and 4). Also, such effects were larger when the stolon internodes were longer or when the leaves were present (Fig. 5c–l), suggesting that the effects increased with increasing the total amount of the reserves. Of the 840 ramets attached with either a proximal internode or both a proximal and a distal internode, 25% still had a visible, healthy proximal internode and 75% of the proximal internodes were depleted completely at harvest; in contrast, of the 840 ramets attached with either a distal internode or both, only five (0.6%) still had a visible, healthy distal internode at harvest (Dong and Yu unpubl. data.). One explanation for the observed position effects on plant growth is, therefore, that the carbohydrate and nitrogen reserves in the proximal internodes could not be used as efficiently by the juvenile, unrooted ramets of *A. philoxeroides* as those in the distal internodes, and some reserves in the proximal internodes could not be readily mobilized to the ramets. Previous studies have shown that structural constraints such as sectoriality and physiological constraints such as formation of discrete integrated

physiological units (IPUs) can confine the mobilization of carbohydrates and nutrients within a clone (Watson and Casper 1984; Mashall and Price 1997). Therefore, there may be larger structural or physiological constraints between the ramet (node) and its attached proximal internode, which restricted the mobilization of the carbohydrates and soluble proteins stored in the proximal internodes so that they could not be efficiently used for the growth of the ramets. Further studies that compare in detail the anatomical structure of the stolon internodes at different parts (i.e. proximal and distal) and trace carbon and nitrogen movements along the stolons of *A. philoxeroides* may help to elucidate the exact mechanisms behind.

Another possible explanation for the observed position effects on plant growth is that carbohydrate support from stolon internodes was important only at the beginning of regrowth of the plants (fragments; Richards and Caldwell 1985). When new ramets (new leaves and roots) were produced, the plants did not continue to depend on storage but could attain sufficient resources through assimilation by new regenerated tissues. At that time older tissues started to bring more costs than benefits because their maintenance needed carbohydrates (for respiration). Given that all the distal internodes were already dead (i.e. completely depleted) at the early stage of the experiment (i.e. within 3 weeks), presence of proximal internodes would greatly reduce the growth of the plants at the time of harvest (at week 9). Contrary to growth, survival of ramets attached with a distal internode had a lower survival rate (Fig. 3a).

We conclude that in frequently disturbed habitats such as rivers, ditches, lawns, croplands and roadsides, where *A. philoxeroides* is distributed, juvenile, unrooted ramets of *A. philoxeroides* can use the reserves in their attached internodes and leaves to regenerate after clone fragmentation. This strategy may contribute greatly to the invasiveness of *A. philoxeroides*, and facilitate its invasion into frequently disturbed, local communities.

Acknowledgments We thank Lei Ning, Wei Wu and Huan Li for assistance with measurements and harvest, Prof. André O. Simões (Universidade de São Paulo) for translation of a paper from Portuguese into English, and two anonymous reviewers for valuable comments on an early version of the manuscript. This research is supported by the Distinguished Scholar Project of Beijing Forestry University (BLJC200910).

References

- Alpert P (1999) Clonal integration in *Fragaria chiloensis* differs between populations: ramets from grassland are selfish. *Oecologia* 120:69–76
- Alpert P, Holzapfel C, Slominski C (2003) Differences in performance between genotypes of *Fragaria chiloensis* with different degrees of resource sharing. *J Ecol* 91:27–35
- Birch CPD, Hutchings MJ (1994) Exploitation of patchily distributed soil resources by the clonal herb *Glechoma hederacea*. *J Ecol* 82:653–664
- Bornette G, Tabacchi E, Hupp C, Puijalon S, Rostan JC (2008) A model of plant strategies in fluvial hydrosystems. *Freshw Biol* 53:1692–1705
- Bouchart V, Macduff JH, Ourry A, Svenning MM, Gay AP, Simon JC et al (1998) Seasonal pattern of accumulation and effects of low temperatures on storage compounds in *Trifolium repens*. *Physiol Plant* 104:65–74
- Corre N, Bouchart V, Ourry A, Boucaud J (1996) Mobilization of nitrogen reserves during regrowth of defoliated *Trifolium repens* L. and identification of potential vegetative storage proteins. *J Exp Bot* 47:1111–1118
- Danckwerts JE, Gordon AJ (1989) Long-term partitioning, storage and remobilization of ¹⁴C assimilated by *Trifolium repens* (cv. Blanc). *Ann Bot* 64:533–544
- Fry JD (1993) Freezing tolerance and carbohydrate content of low-temperature-acclimated and nonacclimated centipede grass. *Crop Sci* 33:1051–1055

- Goulas E, le Dily F, Teissedre L, Corbel G, Robin C, Ourry A (2001) Vegetative storage proteins in white clover (*Trifolium repens* L.): quantitative and qualitative features. *Ann Bot* 88:789–795
- Granstedt RC, Huffaker RC (1982) Identification of the leaf vacuole as a major nitrate storage pool. *Plant Physiol* 70:410–413
- Holm LG, Doll J, Holm E, Pancho J, Herberger J (1997) World weeds: natural histories and distribution. Wiley, New York, pp 37–44
- Jia X, Zhang XZ, Pan XY, Li B, Chen JK (2008) Vegetative propagation characteristics of *Alternanthera philoxeroides* in response to disturbances. *Biodivers Sci* 16:229–235
- Jonsdottir IS, Watson M (1997) Extensive physiological integration: an adaptive trait in resource-poor environments? In: de Kroon H, van Groenendael J (eds) The ecology and evolution of clonal plants. Backbuys Publishers, Leiden, pp 109–136
- Julien MH, Scott JK, Orapa W, Paynter Q (2007) History, opportunities and challenges for biological control in Australia, New Zealand and the Pacific islands. *Crop Prot* 26:255–265
- Klimeš L, Klimešová J, Hendriks R, van Groenendael J (1997) Clonal plant architectures: a comparative analysis of form and function. In: de Kroon H, van Groenendael J (eds) The ecology and evolution of clonal plants. Backhuys Publishers, Leiden, pp 1–29
- Klimešová J, Klimeš L (2003) Resprouting of herbs in disturbed habitats: is it adequately described by Bellingham-Sparrow's model? *Oikos* 103:225–229
- Klimešová J, Kociánová A, Martínková J (2008) Weeds that can do both tricks: vegetative versus generative regeneration of the short-lived root-sprouting herbs *Rorippa palustris* and *Barbarea vulgaris*. *Weed Res* 48:131–135
- Latzel V, Klimešová J (2009) Fitness of resprouters versus seeders in relation to nutrient availability in two *Plantago* species. *Acta Oecol* 35:541–547
- Lawson AR, Kelly KB, Sale PWG (2000) Defoliation frequency and cultivar effects on storage and utilisation of stolon and root in white clover. *Aust J Agric Res* 51:1039–1046
- Ma R, Wang R (2005) Invasive mechanism and biological control of alligatorweed, *Alternanthera philoxeroides* (Amaranthaceae), in China. *Chinese J Appl Environ Biol* 11:246–250
- Marshall C (1990) Source-sink relations of interconnected ramets. In: van Groenendael J, de Kroon H (eds) Clonal growth in plants: regulation and function. SPB Academic Publishing, The Hague, pp 23–41
- Marshall C, Price EAC (1997) Sectoriality and its implications for physiological integration. In: de Kroon H, van Groenendael J (eds) The ecology and evolution of clonal plants. Backbuys Publishers, Leiden, pp 79–107
- Martínková J, Kocvarová M, Klimešová J (2004) Resprouting after disturbance in the short-lived herb *Rorippa palustris* (Brassicaceae): an experiment with juveniles. *Acta Oecol* 25:143–150
- Pan XY, Geng YP, Zhang WJ, Li B, Chen JK (2006) The influence of abiotic stress and phenotypic on the distribution of invasive *Alternanthera philoxeroides* along a riparian zone. *Acta Oecol* 30:333–341
- Pan XY, Geng YP, Sosa A, Zhang WJ, Li B, Chen JK (2007) Invasive *Alternanthera philoxeroides*: biology, ecology and management. *Acta Phytotaxonomica Sin* 45:884–900
- Price EAC, Hutchings MJ (1992) The causes and developmental effects of integration and independence between different parts of *Glechoma hederacea* clones. *Oikos* 63:376–386
- Price EAC, Hutchings MJ, Marshall C (1996) Causes and consequences of sectoriality in the clonal herb *Glechoma hederacea*. *Vegetatio* 127:41–54
- Puijalon S, Piola F, Bornette G (2008) Abiotic stresses increase plant regeneration ability. *Evol Ecol* 22:493–506
- Richards JH, Caldwell MM (1985) Soluble carbohydrates, concurrent photosynthesis and efficiency in regrowth following defoliation: a field study with *Agropyron* species. *J Appl Ecol* 22:907–920
- Roiloa SR, Alpert P, Tharayil N, Hancock G, Bhowmik PC (2007) Greater capacity for division of labour in clones of *Fragaria chiloensis* from patchier habitats. *J Ecol* 95:397–405
- Sainty G, McCorkelle G, Julien M (1998) Control and spread of alligator weed *Alternanthera philoxeroides* (Mart.) Griseb., in Australia: lessons for other regions. *Wetland Ecol Manage* 5:195–201
- Schooler S, Baron Z, Julien M (2006) Effect of simulated and actual herbivory on alligator weed, *Alternanthera philoxeroides*, growth and reproduction. *Biol Control* 36:74–79
- Stuefer JF, Huber H (1999) The role of stolon internodes for ramet survival after clonal fragmentation in *Potentilla anserina*. *Ecol Lett* 2:135–139
- Suzuki JI, Stuefer JE (1999) On the ecological and evolutionary significance of storage in clonal plant. *Plant Species Biol* 14:11–17
- Tao Y, Jiang M (2004) Study on anatomical structure adaptation of stem of *Althernanthera philoxeroides* (Mart.) Griseb to various water conditions. *J Wuhan Bot Res* 22:65–71
- Tao Y, Chen S, Jiang M (2004) Morphological adaptation of *Althernanthera philoxeroides* (Mart.) Griseb to the change of water. *Resour Environ Yangtze Basin* 13:454–459

- Turner LB, Pollock CJ (1998) Changes in stolon carbohydrates during the winter in four varieties of white clover (*Trifolium repens* L.) with contrasting hardiness. *Ann Bot* 81:97–107
- van Kleunen M, Stuefer JF (1999) Quantifying the effects of reciprocal assimilate and water translocation in a clonal plant by the use of stream-girdling. *Oikos* 85:135–145
- Wang BR, Li WG, Wang JB (2005) Genetic diversity of *Alternanthera philoxeroides* in China. *Aquat Bot* 81:277–283
- Wang N, Yu F-H, Li P-X, He W-M, Liu F-H, Liu J-M, Dong M (2008) Clonal integration affects growth, photosynthetic efficiency and biomass allocation, but not the competitive ability, of the alien invasive *Alternanthera philoxeroides* under severe stress. *Ann Bot* 101:671–678
- Watson MA, Casper BB (1984) Morphogenetic constraints on patterns of carbon distribution in plants. *Ann Rev Ecol Syst* 15:233–258
- Wijesinghe DK, Hutchings MJ (1999) The effects of environmental heterogeneity on the performance of *Glechoma hederacea*: the interactions between patch contrast and patch scale. *J Ecol* 87:860–872
- Ye WH, Li J, Cao HL, Ge XJ (2003) Genetic uniformity of *Alternanthera philoxeroides* in South China. *Weed Res* 43:297–302
- Yu F-H, Wang N, Alpert P, He W-M, Dong M (2009) Physiological integration in an introduced, invasive plant increases its spread into experimental communities and modifies their structure. *Am J Bot* 96:1983–1989