



Nutrients and disturbance history in two *Plantago* species: maternal effects as a clue for observed dichotomy between resprouting and seeding strategies

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We assessed the role of nutrients and disturbance experienced by mothers (maternal effects) in the growth of progeny in a pot experiment using two *Plantago* species. Photosynthetic capacity, biomass allocation and fecundity were measured. Offspring of plants grown in nutrient poor conditions produced more leaves, spikes and longer leaves and in case of *P. lanceolata*, they had also higher photosynthetic capacity. The progeny of *P. media* mothers that had resprouted after disturbance was favored in nutrient poor conditions whereas the progeny of undisturbed plants was favored in nutrient rich conditions.

This study demonstrates that maternal effects may play a role in the success of either a seeding or a resprouting strategy in environments with different nutrient availability. Moreover, we showed that alteration of photosynthetic capacity, even during adult stages, is a mechanism through which maternal plants may impact their progeny.

Plant growth strategies are strongly affected by local environmental conditions (Chapin 1980, Miao and Bazzaz 1990, de Groot et al. 2002, James 2008, Suwa and Maherali 2008). Moreover, it is known that the effects of environmental conditions experienced by plant individuals can be extended to their progeny (Miao et al. 1991a, 1991b, Galloway 2001a, 2001b, 2005). These are known as parental effects and some processes they can influence include germination, growth, competitive ability and/or fecundity of offspring.

Parental effects play a significant role primarily in outcrossing species for which the gene movement via pollen is typically greater than through seeds (Galloway 2005). Wind and/or insects may transfer pollen among populations but seed dispersal is largely limited to sites near the maternal plant. This results in individuals that experience an environment similar to their mother's but highly likely not their father's. Thus, maternal effects may remarkably enhance offspring fitness in the maternal environment (Galloway 2005).

Environment-induced maternal effects can be mediated through the embryo via storage reserves, hormones, enzymes, toxins and alike in seeds (Roach and Wulff 1987, Rossiter 1996, Huxman et al. 2001). Those effects are expected to mainly influence germination and growth of seedlings and become less relevant during maturity as other environmental factors outweigh the importance of seed quality (Breen and Richards 2008). Epigenetic factors, which are often initiated by stressful circumstances, may

also affect the progeny phenotype (Rossiter 1996, Bossdorf et al. 2008) due to changes in gene expression that are not conditioned by changes in DNA sequence (Richards 2006, Bird 2007).

Nutrient status is often found to be an important factor contributing to environment-induced maternal effects. Miao et al. (1991a, 1991b) showed that maternal nutrient environment influences the competitive ability of progeny. Likewise, Galloway (2001a) showed a strong effect of maternal nutrient environment on the germination of offspring. In both cases, the growth of descendants of mothers from nutrient rich conditions was enhanced and in the case of *Plantago major* (Miao et al. 1991a, 1991b), the effect lasted for three generations. In their experiments, only plants established from seeds were considered. However, vegetative establishment is also an important strategy in frequently disturbed environments and is known to depend on the nutrient status. Species that regenerate only from seeds (further referred to as seeders in the text) are favored in nutrient rich environments as compared to those regenerating mainly vegetatively after disturbance (further referred to as resprouters), which are favored in nutrient poor conditions (Bloom et al. 1985, Chapin 1991, Iwasa and Kubo 1997, Bellingham and Sparrow 2000, García and Zamora 2003, Buhk et al. 2007, but see Klimešová and Martínková 2004, Clarke et al. 2005, Knox and Clarke 2005). The different performance of resprouters and seeders in differently productive environments has been linked to biomass allocation patterns (Midgley 1996,

Iwasa and Kubo 1997) or ontogeny developmental rate (Buhk et al. 2007) but overall it remains poorly understood.

Severe disturbance as well as nutrient deficiency are stressful factors for plants. To cope with them, plants activate stress-response mechanisms, which may be passed to the offspring generation to alter their growth accordingly. Sultan (1996) reported a better growth of *Polygonum persicaria* progeny whose parents had been stressed by water deficiency. We hypothesise that progeny growth of mothers experiencing two stressful factors (disturbance and nutrient shortage) will be more enhanced than progeny growth of mothers experiencing only one type of stress (disturbance or nutrient shortage). Moreover, since the importance of maternal effects may depend on the environmental conditions experienced by the progeny, we hypothesise that the performance of progeny with different disturbance history and/or nutrient status will be affected differently by the nutrient availability of the progeny environment. Specifically, we expect that enhanced growth will be observed in the same conditions as experienced by mothers, which should result in a preference for a resprouting strategy in nutrient poor environments and for a seeding strategy in nutrient rich habitats.

Maternal effects are known to play a significant role in the growth of progeny of some *Plantago* species, especially in *P. lanceolata*. For this species, several factors of the maternal environment have been shown to affect progeny performance such as nutrient status (Miao et al. 1991a, 1991b), light conditions (van Hinsberg 1998) and temperature (Lacey 1996, Lacey and Herr 2000). In order to elucidate the role of maternal effects in progeny performance of mothers with different disturbance histories we designed a pot experiment with two *Plantago* species (*P. lanceolata* and *P. media*). Both species are facultative resprouters, i.e. regenerate from seeds as well as roots after severe disturbance (Klimešová and Klimeš 2006). They occupy disturbed grasslands, e.g. pastures and meadows of central Europe where they are exposed to trampling as well as different nutrient levels due to the activity of herbivores. We manipulated the level of soil nutrients for the disturbed and undisturbed maternal plants as well as for their offspring. Offspring performance (growth, fecundity and photosynthetic rate) was monitored to assess how the maternal effects influence plants performance.

Material and methods

Species

Plantago lanceolata and *P. media* are both protogynous hemicryptophytes. They produce monopodial, short stems with leaf rosettes and have a tap root, which may be replaced by adventitious roots in older plants. Young plants flower during the first year of life and are self-sterile due to the presence of a single-locus gametophytic self-incompatibility system (Ross 1973). Both species are widespread in temperate Europe. *Plantago lanceolata* is found on a wide range of soil types but is particularly successful on nutrient rich substrates in ruderal grasslands. *Plantago media* usually occurs on oligotrophic dry grasslands (Sagar and Harper

1964). Both species are able to regenerate vegetatively from roots after damage (Klimešová and Klimeš 2006).

Maternal plants

To avoid undesirable carry-over environmental effects on species performance, we used seeds from a commercial production garden where plants had been maintained for over ten years in similar conditions without severe disturbance events or nutrient limitation (Planta Naturalis, Czech Republic).

The experiment was set up in May 2005 in a common garden at the Inst. of Botany AS CR in Třeboň (Czech Republic). Seedlings were planted into pots (15×15×20 cm) that were placed into shallow pits filled with sand to avoid frost damage during overwintering and to maintain homogeneous conditions among individuals. A mixture of commercial soil substrate and sand in a 2:3 ratio was used. We subjected the plants to two levels of nutrients in the soil. The nutrient rich group was fertilized with slow-release fertilizer three times per season by adding 1.2 g N, 0.80 g P, 0.57 g K per plant and season. No fertilizer was added to the nutrient poor group. Fertilization took place on 4 May, 13 July and 21 September 2005. Plants were grown in pots until the following season, which is the time needed for the production of a suitable taproot for a root fragment. In May 2006, we selected 40 plants per nutrient level and species. Half of the plants were allocated to the disturbed group (D) and half to the undisturbed group (U). The plants in the disturbed group consisted only of the taproot of each plant cut into a single 8-cm fragment with the shoot base and apical end removed. These fragments were replanted horizontally into pots and were divided between nutrient poor (D0) and nutrient rich conditions (D1) matching the nutrient conditions they were originally planted in. The plants allocated to the undisturbed group were similarly divided among nutrient poor (U0) or nutrient rich (U1) conditions. Pollination was not controlled and ripe seeds were continuously collected during the 2006 season and used for the establishment of offspring in 2008.

Offspring

On 28 April 2008, the seeds of a subgroup of five randomly selected maternal plants (seeds originated from the season 2006) from every treatment group of both species were chosen. From those seeds, 20 offspring of each maternal plant (five mothers per treatment, 20 maternal plants per species) were grown. The offspring of each maternal plant was allocated into either nutrient rich or nutrient poor conditions. Hence, half of the offspring experienced a nutrient level that differed to that of the maternal plant. Fertilization of the offspring generation took place on 28 April, 27 June and 1 September 2008.

Therefore, the experiment consisted of eight different treatment combinations: offspring of disturbed maternal plants grown in two nutrient levels (D0 and D1 in 2005 and 2006) and subsequently grown in the same or different nutrient level (D00, D01, D10 and D11 in 2008, each level has 50 replicates per species – 10 individuals per mother

plant, five mothers per treatment). The same design was used for the offspring of undisturbed mothers (U00, U01, U10 and U11). The general growing conditions of the offspring were similar to the maternal plants.

Measurements

Seeds produced by all maternal plants were weighed and their germination monitored. Seed weight was estimated from the average weight of 30 seeds (three replicates) for every plant. Germination of those seeds was assessed during three weeks in a germination trial in a climate chamber under controlled conditions (20°C, 75% humidity, 14/10 h light/dark regime).

The following variables were recorded in the offspring individuals: length of the longest leaf and the number of spikes (if present) during the 2008 season (35, 57, 75 and 102 days after sowing) and the number of leaves (35 and 57 days after sowing) until the leaves senesced. In a previous experiment it was shown that the number of seeds was highly correlated with the number of spikes (*P. lanceolata* – $R^2 = 0.7032$, $p < 0.0001$, *P. media* – $R^2 = 0.7009$, $p < 0.0001$, $n = 100$ for each species, Latzel and Klimešová unpubl.) and therefore the number of spikes is considered as a fecundity estimate in this study.

We measured net CO₂ assimilation rate and stomatal conductivity of *P. lanceolata* leaves using a portable photosynthesis system equipped with a standard 6-cm² leaf chamber. Due to the small leaf size of *P. media*, a non-destructive measurement was not possible and thus this species was not measured. We chose mostly overcast days to avoid water stress (stomata closure) and light-induced stress to the photosynthetic apparatus. These measurements were taken on 23 and 24 July 2008 (86 and 87 days after sowing). We randomly selected ten individuals per treatment combination and performed the measurements on a single, fully developed non-senescent intact leaf. The chamber conditions were the following: CO₂ concentration of 370 ppm, leaf temperature of 20.0 ± 0.5°C, air flow rate of 500 μmol s⁻¹ and mean relative humidity of 66%. Saturating photosynthetic active radiation irradiance of 1200 μmol m⁻² s⁻¹ was provided by a built-in LED light source. Each measurement took about 150 s, which allowed for the stabilization of CO₂ exchange. We expressed the

light-saturated photosynthetic rate (i.e. photosynthetic capacity) on a leaf area basis.

Statistical analyses

A repeated measures ANOVA with a full factorial design was used to test the effects of nutrient level experienced by the maternal plants (nutrients 1), nutrient status of the progeny (nutrients 2) and disturbance history on the progeny performance of both species, being the time of the measurements the repeated factor. Analyses were performed for a combined dataset as well as for individual species separately. All variables were log transformed prior to analysis. Despite that five mothers were selected randomly in each treatment line, there was a possibility that the mothers differed from other individuals in the same treatment and the effect could be passed to the progeny. To account for this potential effect of the mother's size on progeny performance (e.g. genetically conditioned size), we calculated the index of similarity for each selected maternal plant, which was used as a covariate in statistics. This index shows how the maternal plant differs from an average size of the treatment group. The index was calculated as the mother's dry biomass divided by the average dry biomass of all plants in the treatment group.

For other, not repeatedly measured variables (i.e. seeds quality and photosynthetic rate) a factorial ANOVA was used. All analyses were performed using the Statistica 8.0 software package.

Results

All undisturbed maternal plants survived and produced seeds. On the contrary, there was some mortality among disturbed plants. Disturbed individuals of *P. lanceolata* species suffered mortality rates of 20% in nutrient poor and 30% in nutrient rich conditions, respectively. In *P. media*, the mortality rate was 10% in nutrient poor and 20% in nutrient rich conditions, respectively.

The higher nutrient level resulted in heavier seeds produced by maternal plants (Table 1). There was no effect of nutrient level on germination. Seed weight and germination did not differ between offspring with different disturbance histories (Table 1).

Table 1. Mean values and SE of seed weight and germination of progeny of both species according to maternal nutrient availability and disturbance history.

	Seed weight (mg)			Germination (%)		
	Nutrients					
	Low	High	p	Low	High	p
<i>P. lanceolata</i>	0.89 ± 0.12	1.31 ± 0.12	0.016	81.6 ± 2.4	80.4 ± 2	0.829
<i>P. media</i>	0.10 ± 0.03	0.30 ± 0.03	0.0001	93.4 ± 2.0	90.5 ± 1.3	0.232
	Disturbance history					
	Disturbed	Undisturbed	p	Disturbed	Undisturbed	p
<i>P. lanceolata</i>	1.09 ± 0.13	1.11 ± 0.11	0.958	78.6 ± 2.1	82.9 ± 2.4	0.185
<i>P. media</i>	0.24 ± 0.03	0.18 ± 0.03	0.140	91.92 ± 1.38	91.93 ± 1.98	0.990

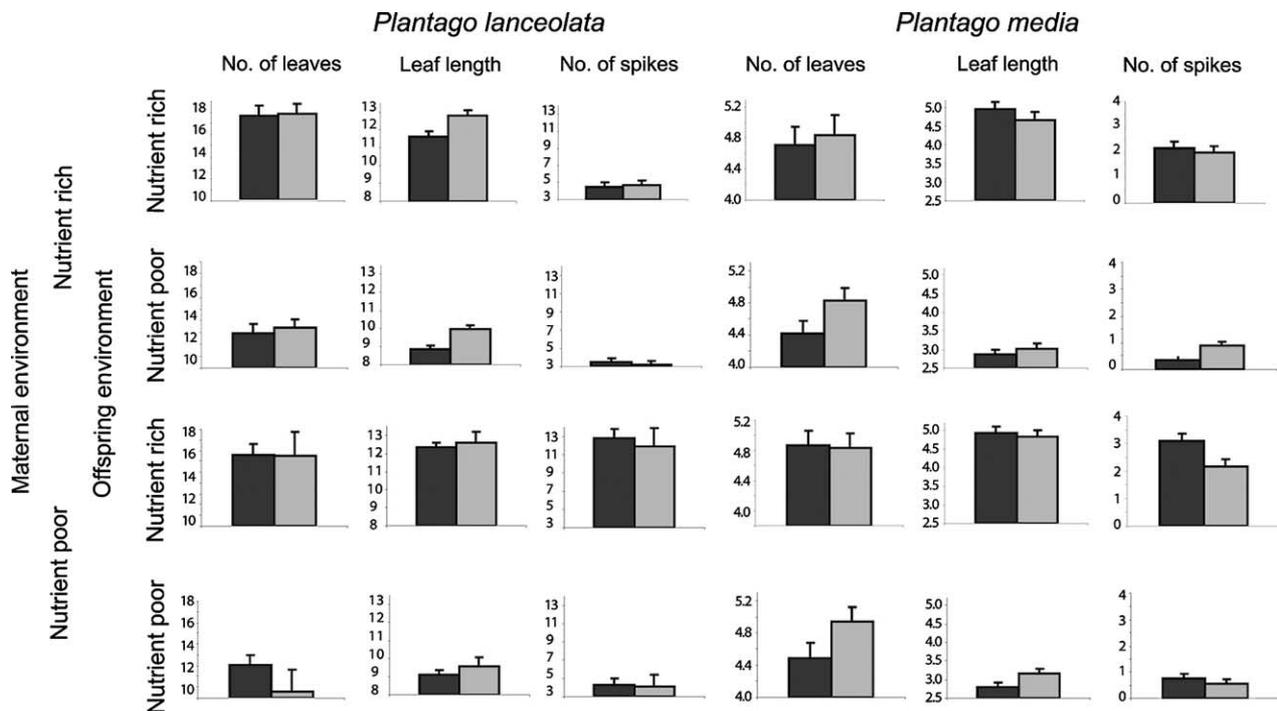


Figure 1. Offspring performance in relation to maternal and offspring nutrient status. Mean values and SE are shown. Grey: offspring of disturbed mothers, black: offspring of undisturbed mothers.

Growth of progeny – cumulative dataset

As expected, species differed in all measured variables (Fig. 1). The nutrient status of the maternal environment (nutrients 1) had a significant effect on the number of leaves, spikes and the length of the longest leaf produced by the progeny (Table 2). Offspring of mothers that experienced nutrient shortage produced more leaves, spikes and longer leaves than those which mothers were grown in a more productive environment. However, the maternal nutrient effect differed between species (interaction $T \times S \times N1$, compare also results for individual species, Table 2). The mean values of all observed parameters are presented in Table 3.

Besides maternal nutrient effects, disturbance experienced by mothers also influenced progeny performance. However, this effect was stronger if the different nutrient conditions in the progeny environment were considered (interaction $N2 \times D$, Table 2, Fig. 2). The offspring grown in the nutrient poor environment produced longer and more leaves if their mothers experienced disturbance, contrary to those individuals whose mothers were undisturbed. The progeny grown under higher nutrient availability showed the opposite response (Fig. 2). Nonetheless, the above mentioned patterns were species specific (interaction $S \times N2 \times D$, compare also results of interactions $N2 \times D$ for *P. lanceolata* and *P. media* separately, Table 2).

Individual species

The progeny of *P. lanceolata* produced significantly more leaves and spikes when their mothers experienced nutrient poor conditions (Table 2, 3). The progeny also produced

longer and more leaves irrespective of the nutrient status of their environment if their mothers were disturbed (Table 2, 3). Nonetheless, they produced longer leaves if their mothers experienced disturbance in the nutrient rich environment whereas leaf length was not affected if their mothers were disturbed in nutrient poor environment (Fig. 3, interaction of $T \times N1 \times D$, Table 2).

Offspring of mothers that experienced low nutrient availability had a higher photosynthetic rate (Table 4, low nutrient availability: mean photosynthetic rate $9.9 \pm 0.42 \mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$; high nutrient availability: mean photosynthetic rate $8.56 \pm 0.41 \mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$). There were, however, no differences in stomatal conductivity, which was measured simultaneously with photosynthesis (Table 4).

The progeny of *P. media* produced slightly but significantly more spikes if the mothers experienced nutrient poor conditions (Table 2, 3). The effect of maternal environment was pronounced during the study and the progeny of mothers from nutrient poor conditions produced more leaves than those of mothers from nutrient rich conditions ($T \times N1$, Table 2).

The progeny of undisturbed mothers stressed by nutrient shortage created more spikes compared to the progeny of disturbed mothers stressed by nutrient shortage. Offspring of undisturbed mothers produced less spikes than offspring of disturbed mothers when the mothers were grown in nutrient rich conditions (interaction $N1 \times D$, Table 2, Fig. 4a). Leaf length and number of leaves and spikes were affected differently by the nutrient status of the progeny and the disturbance history ($N2 \times D$, Table 2, Fig. 4). Differences in leaf length increased during the experiment ($T \times N2 \times D$, Table 2). Progeny of disturbed mothers produced more spikes and more and longer leaves if grown

Table 2. Effect of nutrient level and disturbance history on the performance of progeny of *P. lanceolata* and *P. media*. Results of combined dataset (both species included) and of individual species are presented. Nutrients 1: nutrient level experienced by maternal plants, nutrients 2: nutrient level experienced by offspring. Disturbance history: offspring of disturbed and undisturbed mothers. Statistical significance: *: 0.05 > p > 0.01, **: 0.01 > p > 0.001, ***: 0.001 > p, ns: 0.05 < p. Error of DF is presented in parentheses.

	Combined dataset			<i>Plantago lanceolata</i>			<i>Plantago media</i>		
	Leaf number	Spike number	Leaf length	Leaf number	Spike number	Leaf length	Leaf number	Spike number	Leaf length
	DF 1(784)	DF 1(784)	DF 1(784)	DF 1(392)	DF 1(392)	DF 1(392)	DF 1(392)	DF 1(392)	DF 1(392)
Species (S)	1329.08***	775.82***	3372.58***
Nutrients 1 (N1)	5.65*	19.61***	6.46*	3.96*	22.61***	ns	ns	5.55*	ns
Nutrients 2 (N2)	28.64***	282.12***	220.90***	47.03***	159.86***	188.88***	ns	144.05***	80.97***
Disturbance history (D)	ns	ns	ns	5.56*	ns	11.72***	ns	ns	ns
S×N1	ns	ns	ns
S×N2	19.58***	19.58***	ns
N1×N2	ns	ns	ns	ns	ns	ns	ns	ns	ns
S×D	ns	ns	4.60*
N1×D	ns	ns	ns	ns	ns	ns	ns	5.33*	ns
N2×D	6.03*	ns	6.21*	ns	ns	ns	7.90**	8.27**	6.42*
S×N1×N2	ns	ns	ns
S×N1×D	ns	11.35***	ns
S×N2×D	ns	ns	4.88*
N1×N2×D	ns	ns	ns	ns	ns	ns	ns	ns	ns
S×N1×N2×D	ns	ns	ns
Time (T)	DF 1(784)	DF 2(784)	DF 3(2352)	DF 1(392)	DF 2(392)	DF 3(1176)	DF 1(392)	DF 1(392)	DF 3(1176)
T×S	1929.83***	539.98***	1190.82***	1224.52***	402.02***	160.68***	281.21***	337.75***	944.21***
T×N1	638.39***	21.04***	370.39***
T×N2	ns	ns	ns	7.69**	14.05***	ns	4.71*	4.32*	ns
T×D	ns	157.84***	150.34***	ns	108.43***	65.97***	ns	125.27***	83.09***
T×S×N1	5.15*	ns	ns	ns	ns	ns	5.12*	ns	ns
T×S×N2	8.51**	6.56*	ns
T×S×D	ns	ns	9.55***
T×N1×N2	ns	4.82*	ns	ns	ns	ns	ns	ns	ns
T×S×D	ns	ns	ns
T×N1×D	ns	ns	ns	ns	ns	4.70*	ns	ns	ns
T×N2×D	ns	ns	ns	ns	ns	ns	ns	ns	3.40*
T×S×N1×N2	ns	ns	ns
T×S×N1×D	ns	ns	5.07**
T×S×N2×D	ns	ns	3.12*
T×N1×N2×D	ns	ns	ns	ns	ns	ns	ns	ns	ns
T×S×N1×N2×D	ns	ns	3.59*

Table 3. Mean values and SE of all measured parameters for the different maternal and offspring nutrient status and disturbance history. Nutrients 1: nutrient level experienced by maternal plants, nutrients 2: nutrient level experienced by offspring. Disturbance history: offspring of disturbed and undisturbed mothers.

	Combined dataset						<i>P. lanceolata</i>			<i>P. media</i>		
	Leaf number		Spike number		Leaf length		Leaf number		Spike number		Leaf length	
	Mean	SE	Mean	SE	Mean	SE	Mean	SE	Mean	SE	Mean	SE
Nutrients 1	low	10.98 ± 0.24	9.09 ± 0.47	7.59 ± 0.08	17.08 ± 0.42	10.27 ± 0.31	11.25 ± 0.12	4.88 ± 0.10	3.06 ± 0.17	3.93 ± 0.08		
	high	10.13 ± 0.23	7.98 ± 0.42	7.35 ± 0.07	15.59 ± 0.42	8.81 ± 0.33	10.82 ± 0.12	4.67 ± 0.09	2.94 ± 0.15	3.88 ± 0.08		
Nutrients 2	low	9.38 ± 0.23	4.99 ± 0.53	6.26 ± 0.08	14.12 ± 0.42	5.43 ± 0.34	9.57 ± 0.12	4.64 ± 0.10	1.99 ± 0.19	2.95 ± 0.08		
	high	11.73 ± 0.23	12.09 ± 0.36	8.68 ± 0.08	18.55 ± 0.42	13.65 ± 0.30	12.50 ± 0.12	4.92 ± 0.10	4.01 ± 0.12	4.86 ± 0.08		
Disturbance history	undisturbed	10.14 ± 0.24	8.26 ± 0.48	7.30 ± 0.08	15.62 ± 0.42	9.16 ± 0.31	10.74 ± 0.12	4.65 ± 0.10	3.02 ± 0.17	3.87 ± 0.08		
	disturbed	10.97 ± 0.23	8.81 ± 0.42	7.64 ± 0.08	17.05 ± 0.42	9.92 ± 0.33	11.34 ± 0.12	4.90 ± 0.10	2.98 ± 0.15	3.94 ± 0.08		

in nutrient poor conditions compared to offspring of undisturbed mothers. The opposite response was observed in nutrient rich conditions (N2 × D).

Discussion

Our study showed that the progeny of mothers that experienced both stressful factors (nutrient shortage and disturbance) was not affected more than progeny of mothers that experienced only one type of stress. It is probable that maternal effects induced by stressful factors are not affecting the growth of the progeny in an additive manner. Nonetheless, the performance of progeny was differently affected by the nutrient status of their environment depending on the disturbance history (whether their mothers were disturbed or not). Despite that an additive effect of two stressful factors was not confirmed in our study, it is probable that maternal effects due to disturbance could alter success of the respective regenerative strategies in differently productive environments. Offspring of undisturbed mothers of *P. media* were heavily disadvantaged on low nutrient soils producing fewer, smaller leaves and fewer seeds, whereas offspring of resprouted mothers were little affected by the nutrient levels. Thus, over time in low nutrient and frequently disturbed environments, repeated occurrence of this pattern would tend to select for a resprouting strategy over the seeder strategy due to maternal effects.

Our study demonstrated that the progeny of mothers that experienced nutrient shortage showed better growth both in nutrient poor maternal environment, as well as in nutrient rich, non-maternal environment. This finding is in accordance with an study showing that an stressful maternal environment results in enhanced growth of progeny (Sultan 1996). However, a couple of studies have demonstrated that maternal (parental) effects are responsible for enhanced growth of progeny in maternal conditions (Donohue and Schmitt 1998, Galloway 2005). The contradictions indicate that the results of similar experiments depend on the context of a study, e.g. position on environmental gradient in relation to growth optimum of the studied species, time-scale evaluated, type of environmental factors and their combinations, measured characteristics etc. (Miao et al. 1991a, 1991b, Lacey 1996, Lacey and Herr 2000, Galloway 2005, Galloway and Etterson 2009). All these factors should be considered in future studies.

The role of disturbance history

The growth and fecundity of the offspring, especially of *P. media*, were affected differently by the disturbance history of the maternal plants and by the nutrient availability in both the maternal and progeny environment. However, the growth and fecundity of offspring from disturbed mothers were less affected by nutrient availability (both by maternal as well as offspring nutrient status) compared to offspring from undisturbed plants. This suggests that the weaker nutrient effect on the performance of offspring of disturbed mothers could be due to disturbance maternal effects and/or genotype choice since not all maternal plants regenerated. Thus, only individuals

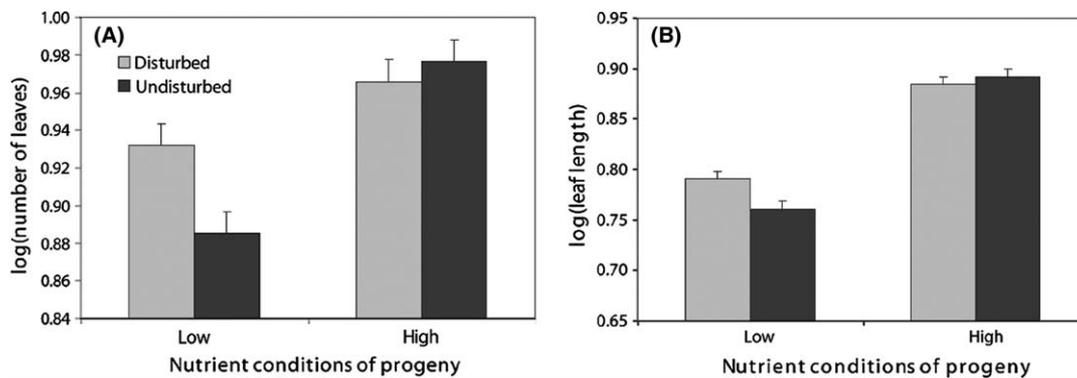


Figure 2. Leaf number (A) and leaf length (B) of progeny of both species according to disturbance history and nutrient status of progeny environment (interaction nutrients $2 \times$ disturbance history, for significance test see Table 2). Low/high: nutrient level experienced by offspring. Mean values and SE are shown.

possessing certain traits which promote resprouting, and probably also other growth characteristics, were able to reproduce, making both genotype and maternal effects potentially important factors affecting growth. Nevertheless, the fact that their fecundity was affected in a different way by maternal and progeny nutrient availability (Fig. 4) suggests that spike production was influenced more by maternal effects than genotypic selection. If selection of maternal plants resulted in offspring with better characteristics in nutrient rich conditions (80% regeneration in disturbed mothers) and played a more important role than maternal effects, then this effect should be apparent also in their performance under higher nutrient availability. However, this was not the case since the progeny of undisturbed plants had higher fecundity than offspring of disturbed

mothers in nutrient rich conditions. Therefore, we conclude that the performance of offspring from mothers with different disturbance history is better explained by maternal effects. Thus, maternal effects could contribute to dichotomy between regenerative strategies as was observed in some woody communities (Bloom et al. 1985, Chapin 1991, Iwasa and Kubo 1997, Bellingham and Sparrow 2000, Buhk et al. 2007). However, this holds truth only if the nutrient conditions of the progeny and disturbance history is considered. If only maternal nutrient environment is taken into account, the opposite effect is observed on the performance of *P. media* plants with different disturbance histories. It is likely that maternal effects due to nutrients and disturbance impact the offspring performance in different directions.

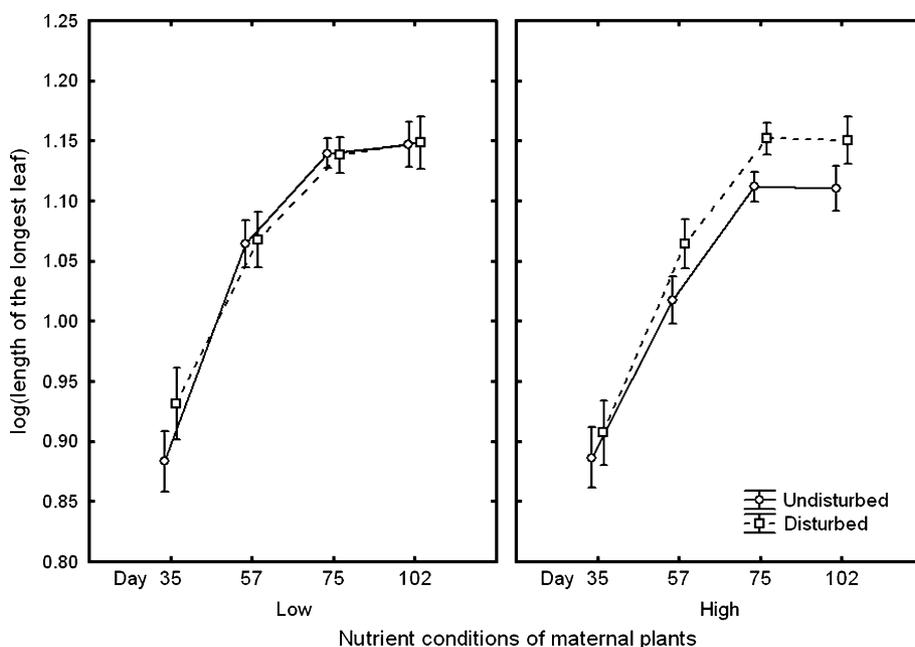


Figure 3. The length of the longest leaf produced by *P. lanceolata* progeny in relation to the nutrient status of maternal plants, disturbance and the day of measurements (interaction time \times nutrients $1 \times$ disturbance history, for significance test see Table 2). Low/high: nutrient level experienced by mothers. Mean values and SE are shown.

Table 4. Effect of nutrient level and disturbance history on photosynthetic capacity and stomatal conductivity of *P. lanceolata* progeny. Day: day of measurement (measured two days), nutrients 1: nutrient level experienced by maternal plants, nutrients 2: nutrient level experienced by offspring, disturbance history: offspring of disturbed or undisturbed mothers. Interactions between day of measurement and other parameters were not significant and thus are not presented in the table. Statistical significance: *: 0.05 > p > 0.01, **: 0.01 > p > 0.001, ***: 0.001 > p, ns: 0.05 > p. DF = 1 and error of DF = 64.

	Photosynthesis	Conductivity
	F	F
Day	17.93***	ns
Nutrients 1 (N1)	5.18*	ns
Nutrients 2 (N2)	15.54***	ns
Disturbance history (D)	ns	ns
N1 × N2	ns	ns
N1 × D	ns	ns
N2 × D	ns	ns
N1 × N2 × D	ns	ns

Mechanisms mediating maternal effects

In the case of *P. lanceolata*, the progeny of mothers grown under nutrient shortage was characterized by a higher photosynthetic rate and production of leaves and spikes. A possible explanation for such phenomenon is that maternal plants may have altered their photosynthetic apparatus to reach higher photosynthetic capacity and photosynthesis nutrient use efficiency in response to nutrient shortage, and their phenotype was probably passed on to their offspring. The effect on photosynthesis might be mediated by enhanced resource acquisition, by alteration of resource allocation towards the photosynthetic apparatus rather than to other functions or by directly influencing the photosynthetic apparatus, for example by some stress induced mechanisms, e.g. genome methylations. The lack of differences in stomatal conductivity suggests that the differences in the photosynthetic rate might be attributed to biochemical rather than stomatal adjustments.

Huxman et al. (2001) also showed offspring photosynthesis adjustments mediated by parental plants which were linked to changes in adult-seed-seedling nitrogen dynamics. They showed that seedlings of parents that were

grown in a CO₂-elevated environment had reduced photosynthesis as compared to seedlings of parents grown in ambient CO₂ conditions. These results suggest that photosynthesis adjustment might be a strategy used by plants to improve the performance of their progeny.

Resource storage in seeds has also been proposed as a mechanism responsible for maternal effects (Roach and Wulff 1987, Rossiter 1996, Huxman et al. 2001). However, this is not likely to have played a role in our study. Our results showed that seed weight, and thus the storage content, were positively affected by higher nutrient levels. However, this did not have any effect on germination or growth of the progeny. Moreover, photosynthesis of *P. lanceolata* was related to the maternal environment and detectable in adult plants when seed storage was depleted. Lacey and Herr (2000) also showed that parental effects influencing *P. lanceolata* progeny performance were independent of seed mass. Miao et al. (1991a) also showed an effect of maternal nutrient conditions on mature progeny of *Plantago* species. In that study, the effect was detectable even in the third generation (Miao et al. 1991b). Thus, epigenetics and/or inherited resource allocation patterns toward photosynthesis might be responsible for the observed results.

Ecological implications of maternal effects

The maternal effects observed in our study may contribute not only to a dichotomy of regenerative strategies in different nutrient conditions but may also result in the evolution of locally adapted populations. As shown in this study, offspring of maternal plants grown in nutrient limited conditions possessed higher photosynthetic capacity and produced more leaves and spikes. Offspring of mothers of *P. media* that varied in their regeneration origin differed in their growth according to the nutrient status of the maternal and/or offspring environment. Individuals from environments with different nutrient conditions will be probably disadvantaged and outcompeted by native plants in stressful, nutrient poor conditions. This could result in preference for maternal ecotypes in maternal environments despite the absence of geographical barriers and the existence of gene flow via pollen. Additionally, progeny from low nutrient environments should be favored in

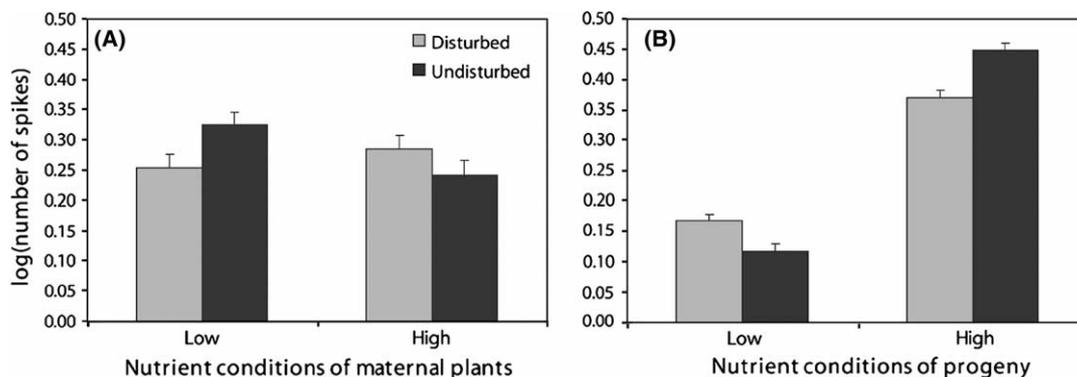


Figure 4. Spike number produced by *P. media* progeny in relation to nutrient status and disturbance experienced by maternal plants (A) and progeny (B). Low/high: nutrient level experienced by mothers. Mean values and SE are shown.

surrounding, more productive environments for at least one generation due to maternal effects. This implies that maternal effects may significantly contribute to population dynamics. This contradicts the conservative opinion that the evolution of locally adapted populations requires consistent geographic variation in selection regimes that cause directional trait changes, as well as limited gene flow among populations. Selective pressures that can result in locally specialized ecotypes are considered to be driven mainly by heterogeneity in abiotic factors such as climate (Joshi et al. 2001, McKay et al. 2005), soil conditions (Snaydon and Davies 1982, Gauthier et al. 1998), or by biotic factors such as competitors, parasites or pathogens (Parker 1995, Linhart and Grant 1996, Gilbert 2002). Our study provides evidence supporting theoretical models that predict a significant role of maternal effects in the rate and direction of evolutionary change in response to directional selection (Riska 1989, Wade 1998, Räsänen and Kruuk 2007), along with an effect on dispersal, gene flow (Donohue 1999, Hereford and Moriuchi 2005) and population dynamics (Benton et al. 2005, Plaistow et al. 2006).

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