

Root proliferation and seed yield in response to spatial heterogeneity of below-ground competition

Erin E. O'Brien¹, Mordechai Gersani² and Joel S. Brown³

¹Biological Sciences, Ecology, Evolution & Marine Biology, University of California, Santa Barbara, CA 93106, USA; ²Department of Life Sciences, Ben-Gurion University of the Negev, Beer Sheva, 84105 Israel; ³Department of Biological Sciences, University of Illinois at Chicago, 845 W. Taylor St., Chicago, IL 60607, USA

Summary

Author for correspondence:
Erin E. O'Brien
Tel: +1 (805) 893 2975
Fax: +1 (805) 893 4724
Email: eobrien@lifesci.ucsb.edu

Received: 24 March 2005
Accepted: 13 June 2005

- Here, we tested the predictions of a 'tragedy of the commons' model of below-ground plant competition in annual plants that experience spatial heterogeneity in their competitive environment. Under interplant competition, the model predicts that a plant should over-proliferate roots relative to what would maximize the collective yield of the plants. We predict that a plant will tailor its root proliferation to local patch conditions, restraining root production when alone and over-proliferating in the presence of other plants.
- A series of experiments were conducted using pairs of pea (*Pisum sativum*) plants occupying two or three pots in which the presence or absence of interplant root competition was varied while nutrient availability per plant was held constant.
- In two-pot experiments, competing plants produced more root mass and less pod mass per individual than plants grown in isolation. In three-pot experiments, peas modulated this response to conditions at the scale of individual pots. Root proliferation in the shared pot was higher compared with the exclusively occupied pot.
- Plants appear to display sophisticated nutrient foraging with outcomes that permit insights into interplant competition.

Key words: below-ground competition, competition, game theory, heterogeneity, pea (*Pisum sativum*), root proliferation, tragedy of the commons.

New Phytologist (2005) doi: 10.1111/j.1469-8137.2005.01520.x

© *New Phytologist* (2005)

Introduction

In addition to responding to nutrients in a heterogeneous environment, plants can respond to the presence of neighboring plants (Mahall & Callaway, 1991; Gersani *et al.*, 1998; Falik *et al.*, 2003). Neighbors may reduce nutrients and alter nutrient heterogeneity available to a focal plant. Consequently, increasing the density of neighbors (or just their root biomass) can decrease several measures of root growth in some plants, including biomass and length (Gersani *et al.*, 1998; Cahill & Casper, 2000; Volis & Shani, 2000). Thus, decreasing nutrient availability and increasing root competition from the roots of other plants appear to have negative effects on the root growth of a plant. This raises the question: Are these plants responding directly to the competitor, indirectly to changes in resource availability, or both?

Some plants are capable of direct interactions. Species as diverse as pea (*Pisum sativum*) (Falik *et al.*, 2003) and *Ambrosia dumosa* (Mahall & Callaway, 1996) discriminate between their own roots and those of neighbors, using this information to adjust root growth in a manner that minimizes intraplant competition. Other plants employ more aggressive tactics, producing toxic chemicals to alter the rooting strategies of neighbors (Schenk *et al.*, 1999; Inderjit & Weston, 2002; Bais *et al.*, 2003, 2004). Assuming that plants do respond directly to competitors, Gersani *et al.* (2001) modeled root competition as a nutrient foraging game where the success of an individual is influenced simultaneously by its own roots, the roots of others, and nutrient availability. Interactions are referred to as 'games' if there are multiple 'players' whose individual strategy choices (e.g. root growth) influence their own payoffs and the payoffs of others (Hammerstein, 1998).

Building on the work of Gleeson & Fry (1997) with individual plants, Gersani *et al.* (2001) predicted that an annual plant will grow more new roots in areas occupied by the roots of a neighbor relative to areas already occupied by its own roots. This growth difference occurs because some of the increased uptake by new roots comes at the expense of decreased uptake by other roots. 'Stealing' nutrients from oneself is counterproductive, while stealing nutrients from the roots of a neighbor can be adaptive. As a consequence, all else being equal, greater root growth should occur in space shared by two or more plants relative to space exclusively occupied by a single plant. This proliferation of roots in shared space results in a 'tragedy of the commons' (Hardin, 1968; Gersani *et al.*, 2001). In response to interplant competition, plants produce more roots and less seed yield than they would if both plants restrained their root production to match resource availability (all else being equal).

The tragedy of the commons theory was originally outlined by Hardin (1968), who proposed that legislation is needed to protect the common good via mutual coercion (Hardin, 1968, 1998). Specifically, he predicted the over-utilization of shared, or public, resources resulting in reduced profits to society at large. When resources are shared, all profits belong to the individual but the costs are shared. This essentially discounts shared resources relative to equivalent but privately held resources (pasture, fish stocks, etc.) resulting in unsustainable harvesting of these resources and reduced total profits in the long term. Hardin (1968, 1998) applied this scenario to several aspects of humanity but never took the leap to other organisms. However, since then, biologists and plant breeders have shown that this idea applies to above-ground tissue production in plants (Poehlman, 1986; Schieving & Poorter, 1999).

The theory applies to root biomass production in annuals as well. Gersani *et al.* (2001) restrict the predictions to annuals because perennials experience an iterated game which allows for a greater variety of root growth strategies. In a two-plant, two-pot experimental system, soybeans (*Glycine max*) produced 85–90% more roots and 30–35% less seed yield when competing (two plants with roots in each of two pots) than when grown in isolation (two plants each with its own pot) (Gersani *et al.*, 2001). In a similar experiment, Kenya beans (*Phaseolus variegatus* var. Kenya) produced 150% more root when competing and 90% more pod biomass when isolated (Maina *et al.*, 2002). These studies do not address how plants allocate roots to patches in a heterogeneous environment caused by varying levels of competition. Given that such heterogeneities are often encountered by plants in nature, it is important to determine if they alter root allocation patterns in response to competition.

Here, using garden peas (*Pisum sativum*), we had two primary goals and several secondary objectives. Our first primary goal was to repeat the two-plant, two-pot experiments of Gersani *et al.* (2001) and Maina *et al.* (2002) to determine whether competing peas give similar results. In such experiments,

a plant is exposed to interplant competition either everywhere or nowhere. *Prediction: two annual plants sharing two pots will produce more roots per individual (and pot) than two plants each with their own pot; two annual plants sharing two pots will produce less yield (in terms of seed number, seed quality or seed biomass) per plant than two plants each with their own pot.*

Our second primary goal was to determine whether an individual plant can modulate its response to spatial heterogeneity in interplant root competition. Plants may occupy multiple patches that vary in intensity of competition. In one study, plants increasingly shifted root allocation away from increased competition intensity in an experiment where an individual had access to two pots varying in the number of competitor plants (Gersani *et al.*, 1998). We adapted the design of Gersani *et al.* (2001) and provided plants with the option of avoiding interplant competition by growing pairs of plants in a series of three connected pots.

The three-pot design allows us to determine whether plants can modulate their rooting patterns in response to local conditions that vary in the presence and absence of interplant competition. The three-pot design also allows us to determine if this localized interplant root competition still results in a reduction of seed yield compared with a situation where each plant has exclusive use of 1.5 pots. This design also allowed us to examine how the root proliferation of an individual in one space influences or is influenced by its root proliferation elsewhere. Such connections between the root growth of a plant in different spaces reveal constraints on total root production and how the costs of producing roots may vary with total root proliferation.

We allowed two competing plants to occupy a shared pot (interplant competition) and an exclusively occupied pot (no interplant competition) within the pot triplet. Alternatively, two noncompeting plants had exclusive use of 1.5 pots each (the 'shared' pot of the competition treatment had a divider preventing interplant competition).

If the plants are able to distinguish the shared pot from their exclusive pot, they are predicted to tailor their root allocation response to the conditions in each pot. *Prediction: total root biomass in the shared pot will be higher than that in a simultaneously occupied exclusive pot.*

When pairs of plants are grown in three pots such that each has exclusive access to one and shares a second with its neighbor, the two plants engage in a two-player nutrient foraging game in the shared pot. The one-player version of this game would be to divide the shared pot in half and give each plant 1.5 exclusive pots. If space and nutrients scale equally, then the plant is predicted to produce half as much roots in its half pot as it does in its full pot. *Prediction: if the shared patch is split in half so that each plant now has exclusive space in half the pot, then root production per plant in the 'shared' pot will be less than when the 'shared' pot is undivided; also, root proliferation will scale linearly with space (pot volume).*

In the three-pot experiment, as in the two-pot experiments, we predict a reduction in reproductive output as a result of the

additional root biomass in the shared pots. As half of the soil environment of a *competing* plant consists of an exclusive pot, the reduction may not be as severe as for plants that *only* have access to shared pots if plants are able to adjust their root allocation to local conditions. *Prediction: two annual plants sharing a pot will produce less yield (in terms of pod or seed number, quality or biomass) per plant than two plants that have access to a split shared pot.*

Our secondary objectives included determining whether the propensity of the plants to engage in nutrient foraging games changes with and is robust to: (1) the initial number of seedling roots per pot; (2) the proximity of neighboring roots to each other; (3) space vs nutrients as the limiting resource, and (4) how nutrients are supplied (pulsed vs continuous delivery).

Methods

Seedling preparation and cultivation

Pea (*Pisum sativum* L.) plants were germinated and their roots split using the protocol of Gersani *et al.* (1998). Trimming the emergent (< 5-mm-long) taproot produced plants with two to four root systems. As the experiments only required plants with two or three root systems, any 'extra' taproots were removed along with all lateral roots. The seedlings were paired by size (< 3 mm difference in root length). They were then transplanted into vermiculite-filled pots. Pots were either 3-inch round standard pots or square pots with equivalent volumes (both 500 ml). Plants were typically not root-bound at the end of any of the experiments, with a few minor exceptions. Any smaller pots used were half the volume (250 ml) of the larger pots.

In the first experiment, to minimize light competition, plants were supported on dowels that ensured straight vertical growth and pairs of pots were closely packed together such that light competition was independent of below-ground competition treatment. In the three-pot experiments, the shoots of different individuals were directed apart so as to prevent any interplant shading. Pairs were arranged into blocks containing a single replicate of each experimental treatment. Nutrients were supplied using various concentrations of Hoagland's solution (Hoagland & Arnon, 1950) via a drip irrigation system at a supply rate of approx. 1 l h⁻¹ per full-sized pot (irrigation events averaged 5 min). The concentrations were varied across experiments to test for the robustness of the root proliferation response to different nutrient concentrations.

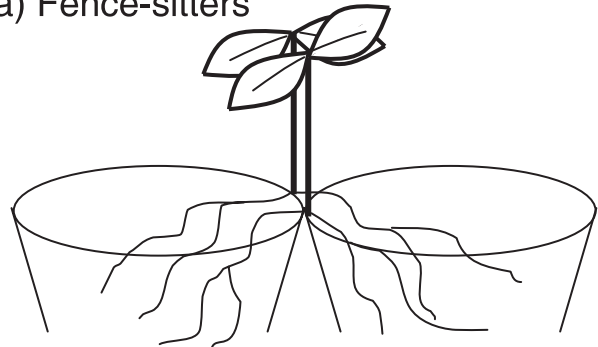
We used root biomass as a measure for quantifying foraging effort in nutrient patches. There are other metrics that could provide additional measures of foraging effort; however, biomass is a simple metric that has successfully revealed treatment effects in similar experiments (Gersani *et al.*, 1998, 2001; Maina *et al.*, 2002).

The two-pot experiment

In this design, our basic experimental unit was a pair of adjacent pots and a pair of pea plants (*Pisum sativum* var. Alaska). This experiment was run in the spring of 1995 at a glasshouse of Ben-Gurion University, Beer Sheva, Israel. Treatments included: (1) each of the two roots of two split-root plants were planted in separate adjacent pots ('fence-sitters' in Fig. 1); (2) each of the two roots of a plant were planted in one pot. A second plant was planted in an adjacent pot ('owners' in Fig. 1). There were 20 replicates arranged in blocks to control for variation in glasshouse conditions (one was discarded because of plant death). The pots were saturated every 2 d with Hoagland's nutrient solution (Hoagland & Arnon, 1950). Excess solution drained through holes in the base of the pot.

We also applied two levels of nutrient solution. Each pot of a pair received either 0.6 or 0.1 strength Hoagland's solution. These concentrations were chosen as 0.1 strength solution was shown to provide adequate nutrient levels for peas, while 0.6 had been previously shown to produce larger plants than 0.1 strength solution. We used drip irrigation to insure that all pots received approx. 66 ml of solution ($\pm 1-3\%$) per watering. Blocks contained all four combinations of planting

(a) Fence-sitters



(b) Owners

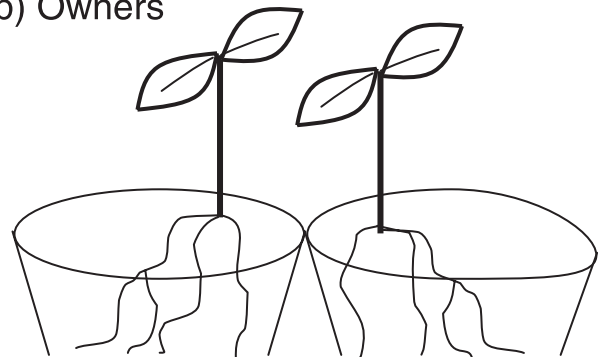


Fig. 1 The two-pot experiment. (a) Fence-sitters: two split-root plants straddled two connected pots, allowing interplant root competition. (b) Owners: two split-root plants each exclusively occupied one of two pots, preventing interplant root competition. Thus, space per individual and the initial numbers of roots per pot were held constant.

treatment (fence-sitter vs owner) and nutrient concentration (0.6 vs 0.1 strength). We harvested the plants after 60 d and measured the dry weight of pods, seeds, leaves, stem and roots.

We used multivariate analyses of variance (MANOVAs) to test for the effects of block, nutrient concentration (0.1 vs 0.6) and planting treatment (fence-sitter vs owner) as well as all two-way interactions on the logarithmic transformations of root mass, pod mass and shoot mass (leaves and stems).

The first three-pot experiment

Using *Pisum sativum* (var. Little Marvel), this experiment was started during the fall of 1997 in the University of Illinois at Chicago temperature-controlled glasshouse (average day temperature 26.6°C; average night temperature 18.3°C; average light ~1200–1500 $\mu\text{mol m}^{-2} \text{s}^{-1}$ with natural sunlight supplemented by fluorescent light to prevent lower light levels) located in Chicago, IL, USA. In this experiment, each of two plants had roots in an exclusive pot and in a shared pot. In the shared pot, we used the presence and absence of a double-layer plastic divider in the shared pot to vary the absence and presence of interplant competition, respectively (Fig. 2; dividers shown in the shared pot in treatments 2, 4 and 6 and in the exclusive pots in treatments 5–6). The dividers were sealed in place with aquarium glue (neutral pH). In these experiments, there were a total of six treatments, half with a divider in the shared pot and three types of plant pairs. Twenty blocks including one replicate of each treatment were used to control for minor differences in glasshouse conditions.

Two planting treatments emerged from varying the initial number of roots in the exclusive pot. Because the initial number of roots of a plant in a given patch could influence final root proliferation, we tested for this by placing either one or two roots of a split-root seedling into the exclusive pot of the plant, representing half or two-thirds of the roots of the seedlings, respectively. In the first case, because each plant needs two roots to occupy the two pots, the exclusive pots started with a total root density that was half that present in the shared pots. In the second case, because we used plants with three roots, it insured that the exclusive pots and the shared pots started with the same root densities. In this treatment an individual plant had two roots in its exclusive pot and one in its shared pot.

A third planting treatments used seedlings with two roots in their exclusive pots plus a divider in the exclusive pot that gave half of the pot to one of the roots of the individual and the other half to the roots of the other plant.

Treatments 1 and 2 (Fig. 2) had plants with two root systems, one in the shared pot and one in the exclusive pot. Treatment 2 (see Fig. 2) was identical except for a divider in the middle of the shared pot which kept the plants from interacting. The comparison of these two treatments examines the ability of plants to control biomass allocation to root systems depending on the presence or absence of interplant root competition.

Treatments 3 and 4 (Fig. 2) determined the influence of the initial numbers of roots on subsequent root total production. Each plant in these treatments had three root systems. Two

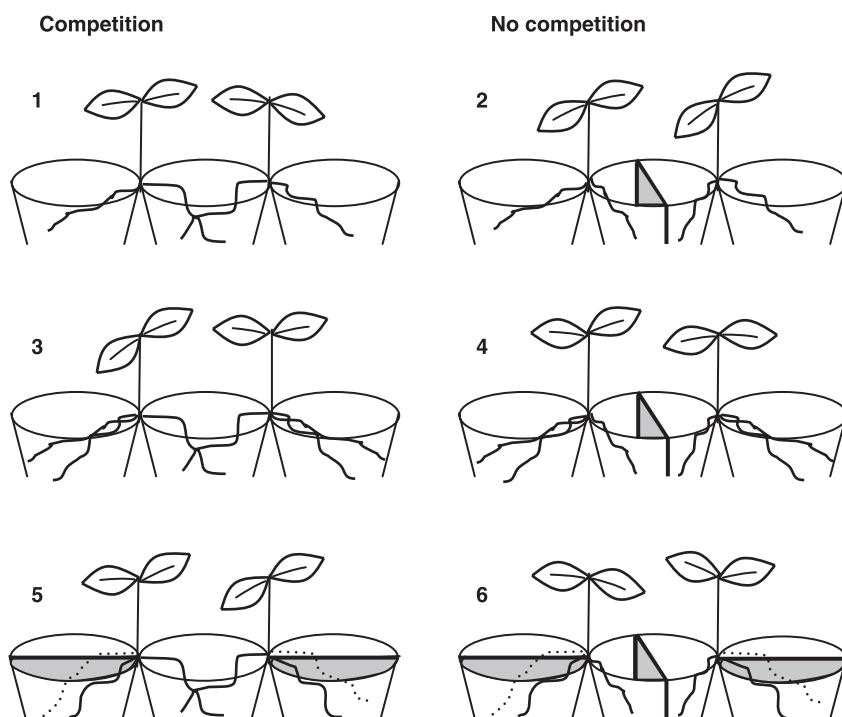


Fig. 2 The first three-pot experiment. Here, two split-root plants shared three pots, with each individual occupying two of the pots. The presence or absence of dividers (shown by a thick line) in the shared pot varied interplant competition (absent or present, respectively). Treatments on the left (1, 3 and 5) permitted interplant competition (two-player game) in the shared pot, while those on the right (2, 4 and 6) had dividers that prevented interplant competition (dividers indicated by gray shading and thick lines). Treatments 3–6 used seedlings with three root systems. Placing two roots of a plant in its exclusive pot kept the initial numbers of roots per pot constant, and tested for the effect of the starting numbers of roots in a pot. A divider between the two roots of a plant in its exclusive pot (treatments 5 and 6) tested for the effect of dividing a pot on root proliferation as well as the effect of intraplant root competition on root proliferation.

roots of a plant were placed in its exclusive pot and its third root in the shared pot. In this way, each pot had equal amounts of roots at the start of the experiment. Not only can the effect of competition be measured by comparing these two treatments, but, by comparing them with treatments 1 and 2, we can determine if there is an effect of initial root number in the exclusive pots.

Treatments 5 and 6 (Fig. 2) also used plants with three root systems and tested for the effect of placing a divider in pots such that the two root systems in the exclusive pots were isolated from each other with a divider similar to the dividers used in the shared pots. Treatment 5 had an undivided shared pot, while treatment 6 had dividers in the shared and exclusive pots. These two treatments show whether the geometry of a split pot influences total root production even when there is no interplant competition.

The seedlings were transplanted into the pot triplets and allowed to grow for 2 months until pods ripened. During this period, they received 50 ml of 0.2 strength Hoagland's solution daily via drip irrigation. This concentration was chosen based on the results of the two-pot experiment. They were also flushed once a week with approx. 250 ml of distilled water.

The pairs of plants were placed in blocks of 12 to factor out the effects of location in the glasshouse. All treatments were present twice in each block, and the duplicates were planted 1 wk apart (the effect of planting time is referred to as 'planting group' henceforth).

At the end of 2 months, the dry weights of pods and seeds, seeds, leaves, stem and root were measured, and the numbers of pods and seeds counted. The biomass data were log-transformed and the count data were square-root-transformed before analysis. We used a MANOVA to test for the main effects of competition (treatments 1, 3 and 5 vs 2, 4 and 6), root number at the start (treatments 1 and 2 vs 3 and 4; start conditions), and splitting of the exclusive pots (treatments 3

and 4 vs 5 and 6; exclusive split) on root biomass in the shared pots, root biomass in the exclusive pots, shoot biomass, pod biomass, seed biomass, pod number, seed number, and seed size. Block and planting group were also included in the analysis as random effects. All measures were from the average of the two plants occupying a given triplet of pots, except for the root biomass in the shared pots which was summed for the two individuals for purposes of comparison with the exclusive pots on a per pot basis.

Nutrient supply experiment (two and three pots)

The two-pot and three-pot experiments outlined above supported the prediction that plants produced more roots in response to interplant than intraplant root competition. As will be seen in the Results section, the shared pots had significantly more root mass than the exclusive pots. To ensure that the result was robust and free of artifacts of the three-pot system, we independently manipulated nutrient regime, pot size, and nutrient level in a series of one-plant, two-pot and two-plant, three-pot treatments (Table 1; Fig. 3).

Three-pot treatments In fall 1998, we repeated the two-plant, three-pot experiment in the Chicago glasshouse with some modifications. In the shared pot of the triplet, the initial roots of the two plants were started 1 cm apart. In the previous experiment, the roots of the two plants in the shared pot were started on opposite sides of the pot, possibly encouraging root over-proliferation as a consequence of initially separate depletion zones around the roots of each seedling.

To test for effects of nutrient supply regime, we ran the three-pot experiments under a relatively continuous nutrient supply regime (irrigated with 400 ml per pot of 0.07 strength Hoagland's solution once a week) vs a more pulsed nutrient supply regime (all 400 ml per pot of 0.35 strength Hoagland's

Table 1 Summary of two- and three-pot treatments in the final experiment (see Fig. 3)

Treatment	Nutrient supply	Pot number and size	Nutrient concentration
a: Two-root plants in competition ¹	Constant	Three full-sized pots	Full strength
b: Two-root plants in competition ¹	Single pulse	Three full-sized pots	Full strength
c: Three-root plants in competition ¹	Constant	Three full-sized pots	Full strength
d: Three-root plants in competition ¹	Single pulse	Three full-sized pots	Full strength
e: Control ²	Constant	Two full-sized pots	Full strength
f: Control ²	Single pulse	Two full-sized pots	Full strength
g: Nutrient reduction ²	Constant	Two full-sized pots	One pot full strength One pot half concentration
h: Nutrient reduction ²	Single pulse	Two full-sized pots	One pot full strength One pot half concentration
i: Space reduction ²	Constant	One full-sized pot One half-sized pot	Full strength
j: Space reduction ²	Single pulse	One full-sized pot One half-sized pot	Full strength

¹Tests for root over-proliferation into a shared pot relative to root growth into exclusive pots for pairs of competing plants.

²Tests for the independent effects of soil volume and total nutrient supply on root proliferation.

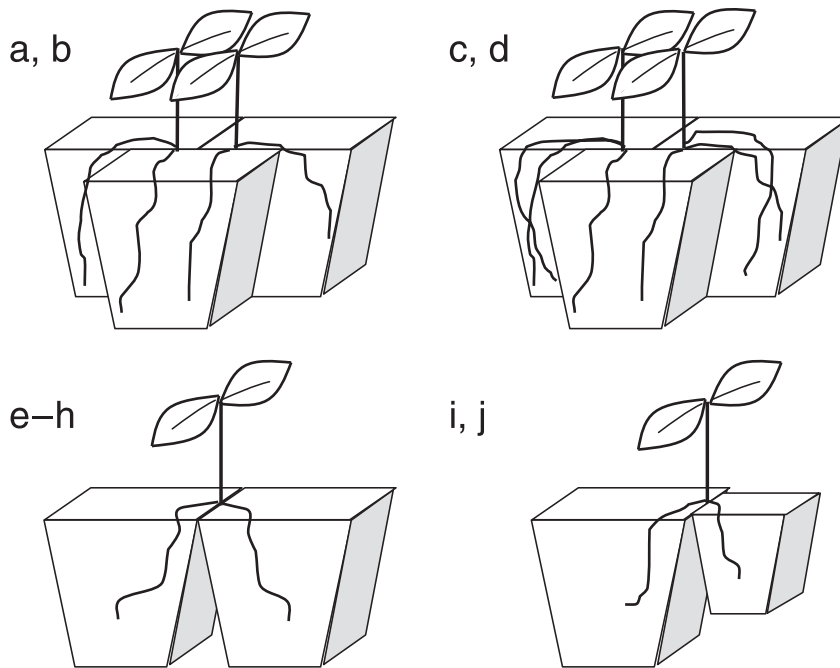


Fig. 3 The nutrient experiment. Treatments a–d repeat the three-pot experiment, except that the roots of the two plants in the shared pot were planted in close initial proximity. Treatments e–j (one plant straddling two pots) test for the various indirect effects of competitors (see Table 1 for nutrient treatments). A, c, e, g and i were provided with all of their nutrients in the initial watering and the remaining treatments received the same total amount of nutrients spread over five waterings to test for the effect of nutrient supply.

solution provided in an initial watering). A volume of 400 ml per pot of de-ionized water was added to the pots every other day, with the exception of nutrient addition days. To test for the effects of initial root numbers, we crossed the nutrient treatments with treatments of one vs two initial roots in the exclusive pot. To do this, we trimmed split-root seedlings to have either two roots (one root per plant in its exclusive pot and one root per plant in the shared pot) or three roots (two roots per plant in its exclusive pot and one root per plant in the shared pot).

Two-pot treatments The over-proliferation of roots in the shared pot may simply be an effect of space, independent of resource availability and/or resource competition. Finally, the architecture of space may influence root proliferation and distort our ability in these experiments to detect responses to shared space vs exclusively occupied space that has been created by splitting pots in half. The model assumes that nutrient availability drives root proliferation, and space matters only insofar as resource density (nutrients per unit volume) and the encounter probability of a root with resources will scale with soil volume. For instance, doubling the amount of space while holding nutrient density constant should result in twice as much root proliferation (twice as much nutrients). To test for space as a separate factor influencing root proliferation, we independently varied pot size and total nutrients per pot.

Treatments consisted of individual plants grown in pairs of pots (Fig. 3; Table 1). In each of the pot pairs, one of the pots was identical in size, shape and nutrient availability to the exclusive pots used in the three-pot experiment described above. In treatments e and f (Fig. 3), the second pot was

identical to the first. In treatments g and h (Fig. 3), the second pot was the same size as the first, but the nutrients were supplied to it at half the concentration supplied to the first pot (same volume of space with half the total amount of nutrients). This treatment would mimic simple competition if each plant occupies the entirety of the shared pot but experiences only half the nutrients by virtue of symmetric competition. In treatments i and j (Fig. 3), the second pot was half the volume of the first pot and received half the volume of full-concentration nutrient solution (half the amount of nutrients but the same nutrient density per unit volume). This treatment would mimic simple competition if each plant occupies just half of the space of the shared pot while experiencing the entirety of nutrients in that half. Because there is no actual interplant competition, we expect the pot receiving half of the nutrients (whether by halving the concentration or by halving the space) to result in approximately half the root proliferation of the control pot. Treatments a, c, e, g and i received constant nutrients while treatments b, d, f, h and j received pulsed nutrients.

Twenty blocks including one replicate of each treatment were used to control for minor differences in glasshouse conditions. After 5 wk, the plants were harvested and processed in the same manner as in the previous experiment, except for the absence of yield data (the plants were harvested before pod production). All biomass data were log-transformed before analysis.

Using a MANOVA, the three-pot treatments (a–d in Fig. 3) were compared to the half-volume two-pot treatment (in effect, half a split triplet; i and j in Fig. 3) to test for the effects of competition (interplant vs no interplant root

competition) and resource supply (pulsed vs continuous) on shoot biomass (averaged over the two plants of a pair for treatments a and b; Fig. 3), root biomass per pot for the shared pot (for the half pots, this meant the combined root biomass of the two plants in their respective half pots), and root biomass in the exclusive whole pots (averaged over the two plants of a pair).

The two-pot data were analyzed separately using a MANOVA to test for the effects of pot size, nutrient concentration and nutrient supply rate on the per-pot root biomass and shoot biomass. In addition, the treatment pots from the two-pot treatments were compared with the shared pots from the three-pot treatments to determine if there was a difference in root allocation in response to competition compared with reductions in resources.

Results

The two-pot experiment

Root, shoot and pod biomass were log-transformed and analyzed in a MANOVA for the effects of block, competition treatment, and nutrient level as well as all two-way interactions. In accord with the predictions of the tragedy of the commons model, the two fence-sitters produced, per individual, significantly less pod mass than owners ($F_{1,198} = 7.631$; $P < 0.01$; competition effect): 45% less at low and 51% less at high nutrient concentrations (Fig. 4; competition \times nutrient interaction). Fence-sitters (competing plants) produced significantly more root mass per individual than owners (isolated individuals) ($F_{1,198} = 6.12$; $P < 0.05$; competition effect): 51% more at low and 7% more at high nutrient concentrations, with a significant interaction effect between competition treatment and nutrient concentration ($F_{1,198} = 3.79$; $P = 0.05$; Fig. 4; competition \times nutrient interaction). Fence-sitters also produced significantly more shoot mass ($F_{1,198} = 9.98$; $P < 0.005$; competition effect): 56% more at low and 7% more at high nutrient concentrations, with a significant interaction effect between planting treatment and nutrient concentration ($F_{1,198} = 9.37$; $P < 0.005$; Fig. 4; competition \times nutrient interaction). Root to shoot ratios remained unchanged with planting treatment.

Root mass ($F_{1,198} = 56.75$; $P < 0.001$), pod mass ($F_{1,198} = 11.34$; $P < 0.001$) and shoot mass ($F_{1,198} = 121.41$; $P < 0.001$) all increased with nutrient concentration (Fig. 4; nutrient effect). Shoot mass saw the largest proportional increase while pod mass saw the smallest proportional increase. There was no effect of block.

The three-pot experiment

Root, shoot, pod and seed biomass data were log-transformed and pod and seed number were square-root-transformed and analyzed in a MANOVA for the effects of block, splitting the shared pots (competition), splitting the exclusive pots

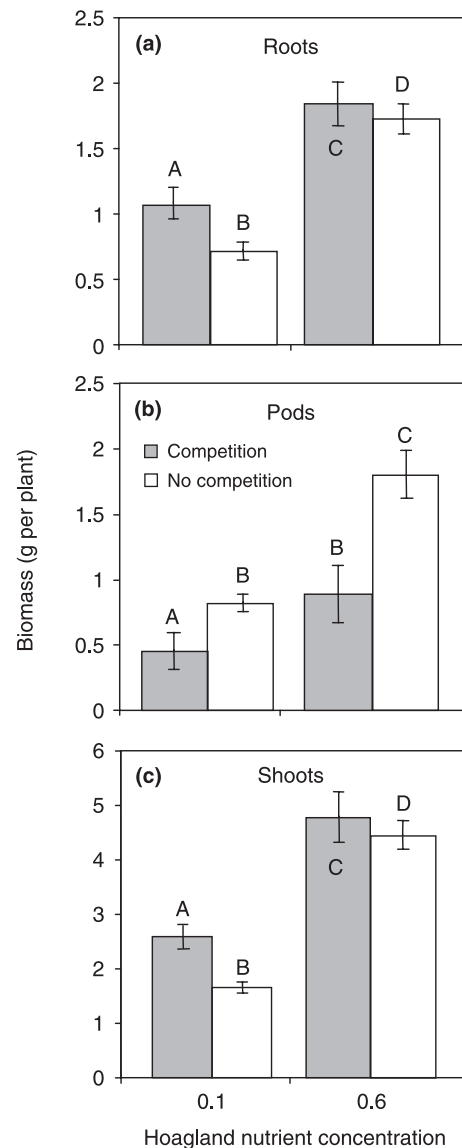


Fig. 4 The effect of nutrients and interplant competition. Fence-sitters (interplant competition) produced significantly more root biomass (a), less pod biomass (b) and more shoot biomass (c) than individuals that did not compete (owners). Nutrient concentration had a significant effect on biomass but did not affect the root over-proliferation response to interplant competition. Bars are standard errors and statistically significant differences are indicated by different uppercase letters (A–D).

(exclusive split), number of roots at the start of the experiment (start conditions), and the planting group (group) as well as all two-way interactions (Table 2). Total root biomass was over 60% higher in the shared pots with interplant competition (treatments 1, 3 and 5; competition effect on roots in the shared pot: $F_{1,57} = 14.216$; $P < 0.0001$; Fig. 5) than in those in which interplant competition was prevented with a divider (treatments 2, 4 and 6). There was the reverse effect of interplant competition on root biomass in the exclusive pots.

Table 2 The multivariate analysis of variance (MANOVA) results for the three-pot competition experiment (see Fig. 2)

Variable	df	Roots: commons		Roots: owned pots		Shoots		Pod mass		Pod number		Seed mass		Seed number		Wilks' lambda	
		MS	F	MS	F	MS	F	MS	F	MS	F	MS	F	MS	F	Value	F
Competition	1	1.714	14.216 ¹	0.334	4.375 ¹	0.005	0.023	0.028	0.404	0.177	0.298	0.002	0.014	0.973	0.337	0.636	4.176 ¹
Start conditions	1	0.184	1.527	0.081	1.056	0.33	1.541	0.054	0.781	2.940	4.952	0.116	0.765	15.546	5.380	0.817	1.628
Split exclusive pots	1	0.246	2.044	0.090	1.184	3.504	16.355 ¹	0.640	9.341 ¹	1.367	2.302	0.774	5.098 ²	8.008	2.771	0.721	2.826 ²
Planting group	1	0.116	0.959	0.006	0.079	0.004	0.020	0.404	5.891	3.414	5.751	1.088	7.161	11.705	4.051	0.831	1.482
Start conditions × competition	1	0.272	2.259	0.307	4.033	1.039	4.847	0.026	0.382	0.244	0.411	0.002	0.013	1.104	0.382	0.879	1.001
Split exclusive × competition	1	0.060	0.497	0.018	0.230	0.275	1.283	0.054	0.795	0.098	0.165	0.151	0.994	0.370	0.128	0.893	0.873
Group × competition	1	0.176	1.461	0.033	0.436	0.038	0.179	0.031	0.449	0.330	0.555	0.054	0.356	4.038	1.397	0.915	0.681
Group × start conditions	1	0.009	0.075	0.014	0.189	0.519	2.424	0.128	1.866	0.710	1.196	0.315	2.075	10.823	3.745	0.855	1.231
Group × split exclusive	1	0.001	0.011	0.006	0.080	0.407	1.901	0.274	3.994	0	0	0.511	3.367	0.580	0.201	0.866	1.132
Error	57	1.089		0.684		1.926		0.621		5.346		1.368		26.01			

We tested for the effect of competition in a shared pot, the effect of initial root distribution (history) and the effect of splitting exclusive pots with dividers on root biomass in shared pots (commons) and in exclusive pots, as well as shoot biomass, pod and seed mass and pod and seed number. Significant *F*-values are indicated by ²*P* < 0.05, ¹*P* < 0.001. df, degrees of freedom; MS, mean square.

Plants experiencing interplant competition in their shared pot (treatments 1, 3 and 5) produced approx. 10% less root in their exclusive pot than those grown separated from their competitors (treatments 2, 4 and 6; $F_{1,57} = 4.375$; $P < 0.05$; Fig. 5; competition effect). There was no significant effect of competition on any other plant measures.

While there was a slight decrease in seed yield for competing plants relative to isolated plants, the difference was not significant (Fig. 5).

The number of root systems a plant started with (one or two in the exclusive pot for a total of two or three per plant; start conditions effect) had no significant effect on final root biomass in either the exclusive or the shared pots, or on any dependent variable. Splitting the exclusive pots with dividers (treatments 5 and 6) caused a small but significant reduction in total shoot biomass, total pod mass and total seed mass (shoot: $F_{1,57} = 16.355$, $P < 0.0001$; pod: $F_{1,57} = 5.098$, $P < 0.05$; seed: $F_{1,57} = 9.341$, $P < 0.005$; exclusive split effect). Splitting the exclusive pots had no effect on root biomass.

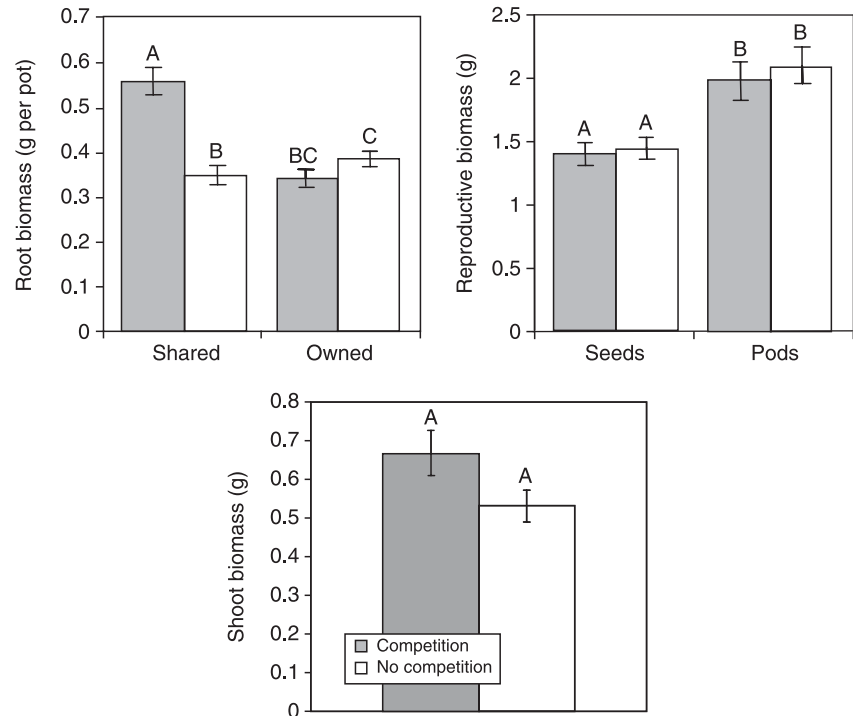
There was no effect of block or planting group on any dependent variable. There were no significant interaction effects among the independent variables.

Two separate and independent statistical tests were also used to look at root biomass production within pairs of plants. We looked for the effect of interplant competition on root production within pairs of plants by comparing total root production in the shared pot (by both plants) with per-pot root production in the exclusive pots of a triplet using a paired *t*-test. There was significantly more root biomass in the shared pots relative to the simultaneously occupied exclusive pots when interplant competition was allowed (treatments 1, 3 and 5; $t_{33} = 6.317$; $P < 0.0001$) and no significant difference when competition was prevented (treatments 2, 4 and 6; $t_{33} = -1.568$; $P < 0.5$). In a second paired *t*-test we compared total root production for pairs of plants per pot type. Total root production in the shared pots (under interplant competition; treatments 1, 3 and 5) was less than the combined root production in the two exclusive pots, indicating that the presence of a competitor did cause an individual to produce fewer roots in the shared pots ($t_{33} = -2.837$; $P < 0.01$). If the plants were completely indifferent to the presence of a competitor, there should have been no difference between the roots a plant produced in an exclusive pot and in a shared pot.

Nutrient supply experiment (two and three pots)

Test for effects of interplant competition The main effects of interplant competition (three-pot treatments vs two-pot treatments with half-volume second pots), number of roots at the start of the experiment (one vs two initial roots in the exclusive pot) and resource availability (continuous vs pulsed) as well as the effect of block on shoot and root biomass (log-transformed) were tested with a MANOVA. The response of plants to interplant competition in the pot triplets showed

Fig. 5 The three-pot experiment. Competing plants produced significantly more root biomass (on a per pot basis) in the shared pot than in an exclusively occupied (exclusive) pot. Pairs of plants separated by a barrier did not display this difference; there was no significant difference in the roots of the shared and exclusive pots (on a per pot basis). The response by competing plants in the shared pot was coupled with a significant reduction in root production in the exclusive pots, suggesting a change in the marginal cost of root production. There were no significant effects of interplant competition on shoot or reproductive biomass per plant. However, the trends in the data accord with the results from the two-pot experiment for reproductive biomass. Bars are standard errors and statistically significant differences are indicated by different uppercase letters (A–C).



that, as in the previous experiments, the total root production in the shared pots was significantly greater than in the two smaller (no competition) pots ($F_{1,50} = 126.413$; $P < 0.001$) regardless of the nutrient treatments.

There was no significant difference between the two-pot control pots (full-volume, full-nutrient pots attached to the half-volume pots) and the three-pot exclusive pots ($F_{1,50} = 0.627$; $P > 0.05$).

Initial root numbers in the exclusive pot (treatments a and b vs c and d; start conditions) did not significantly influence root biomass in either the shared pots ($F_{1,50} = 1.799$; $P > 0.05$) or the exclusive pots ($F_{1,50} = 0.173$; $P > 0.05$), or shoot production ($F_{1,50} = 0.873$; $P > 0.05$). The effect of pulsing the resource may be spurious as the univariate results suggest significant effects (but see separate two-pot analysis below) on root production in the shared ($F_{1,50} = 7.534$; $P < 0.01$) and exclusive pots ($F_{1,50} = 4.655$; $P < 0.05$), as well as on shoot production ($F_{1,50} = 5.151$; $P < 0.05$). However, Wilks' lambda multivariate was 0.865 with a P -value of 0.072. There were no significant interactions between any of the independent variables.

The significant increase of root biomass/pot in shared pots as a result of competition was again verified with a t -test that compared root production per pot in the exclusive pots vs the shared pot when the shared pot was undivided and permitted interplant root competition ($t_{37} = 15.581$; $P < 0.001$).

Two-pot treatments The effect of reduced space and nutrients within each of the two-pot arrangements could also be compared using paired t -tests. There were 41% fewer roots in

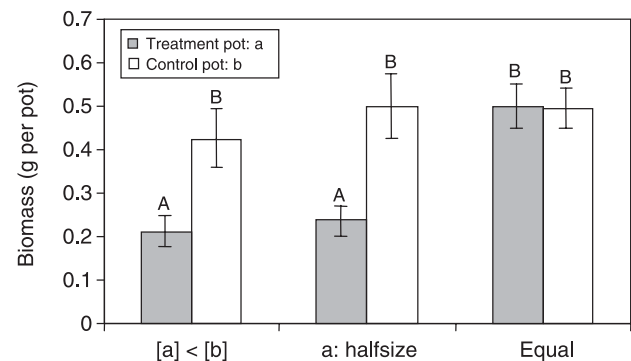


Fig. 6 Effect of pot size and nutrient amount. [a] < [b]: half-nutrient treatment; a: halfsize: half-volume treatment; Equal: pot size and nutrient amount equal to those for the control pot. The two-pot treatments from the nutrient experiment show that halving the space and/or nutrient concentration resulted in a similar and significant decrease in root biomass of c. 50%. This differs from the effect of symmetric competition on root biomass. Bars are standard errors and statistically significant differences are indicated by different uppercase letters (A and B).

the half-nutrient pots compared with the control plants (treatments e and f vs g and h; $t_{14} = 2.993$; $P = 0.01$; Fig. 6) and 46% fewer roots in the half-volume pots compared with the control plants (treatments e and f vs i and j; $t_{17} = 4.373$; $P < 0.001$; Fig. 6).

In a separate MANOVA, we used treatments e through j to determine the effect of reduced space, reduced nutrients and resource supply rate. There was significantly less root mass

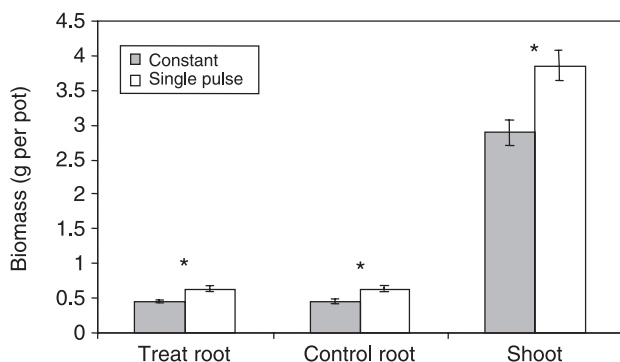


Fig. 7 Nutrient supply regime effects. Plants were generally larger under the single pulse nutrient regime compared with the more constant supply treatments, and this held true for all types of biomass measured. This effect did not interact with plant responses to interplant competition. Bars are standard errors and significant differences are indicated by asterisks.

allocated to the pots that had half of the total nutrients or half the total volume, in accord with the above *t*-tests (see above for treatment-specific amounts; $F_{2,47} = 12.293$; $P < 0.001$; Wilks' lambda = 0.324), but there was no effect of pot size or nutrient concentration in the manipulated pot on the root production of the plant in its control pot. The plants in the pulsed nutrient supply treatments (f, h and j) produced 47% more roots in their control pots ($F_{2,47} = 8.153$; $P < 0.01$; Wilks' lambda = 0.821 applies to all nutrient supply effects; Fig. 7), 53% more roots in their treatment pots ($F_{2,47} = 8.424$; $P < 0.01$; Fig. 7) and 28% more shoot ($F_{2,47} = 4.004$; $P = 0.05$; Fig. 7) than plants in the constant supply (e, g and j). There was not a significant interaction effect between pot treatments and resource availability.

Discussion

We investigated how plants vary their root growth in response to intraplant vs interplant root competition in a variety of environments. In a two-pot experiment, we showed that interplant competition resulted in a 'tragedy of the commons' for peas. Relative to intraplant root competition, competing individuals increased root growth and suffered a concomitant reduction in seed production. Plants produced significantly more roots when faced with a competitor than with a halving of space or nutrients. The three-pot experiments suggest that peas are capable of compartmentalizing their response to various patches, thus maximizing the whole-plant benefits of their root systems. Using three-pot experiments, we showed how an individual pea plant can vary its root growth to local conditions that differ in the presence of the roots of another plant. This localized ability of peas to amplify root production in response to interplant root competition was robust to a wide variety of conditions. The peas, by producing more roots in intact shared pots than when split in half for exclusive use, appear to respond directly to the presence of the competitors

rather than just to the decline of nutrients produced by the roots of the competitor.

Other examples of root over-proliferation can be found in the literature. Some trees and wild strawberries produce more roots when competing compared to when alone (Holzapfel & Alpert, 2003; King, 1993). Day *et al.* (2003) found a root over-proliferation response in a Poaceae species, although a second Poaceae species did not show such a response.

We have also shown that there may be reduced fitness in terms of seed yield for annual plants that do not segregate their root systems and that this reduction is context specific. In contrast to the two-pot experiment, the over-proliferation of roots in the shared pot of the three-pot experiments did not cause a statistically significant reduction in seed production (although the trends were in that direction). Having an exclusive pot may mitigate the negative consequences of interplant competition in the shared pot. As earlier studies of this phenomenon did not simultaneously provide both types of patches to plants, it is not clear how widespread this effect is. Although most plants do not often encounter areas they exclusively occupy, they likely do experience heterogeneity in the competition landscape. It will be important to determine if only exclusive space can ameliorate the effects of root over-proliferation or if other types of patches may have a similar effect, and if other species display a similar strategy under these conditions.

The tendency for plants to suffer a reduction in yield from over-proliferating roots in response to interplant competition may or may not be widespread in nature. Three agricultural species have now been shown to exhibit an increase in root production and a decrease in seed yield in response to interplant competition (soybean: Gersani *et al.*, 2001; Kenya beans: Maina *et al.*, 2002; pea: this study). The discovery of a similar reduction in yield as a result of over-production of above-ground tissue (plant height) led to some of the significant increases in yield during the green revolution as agronomists bred for shorter strains of many plants (Poehlman, 1987). Through the intervention of artificial selection, plants reallocated resources, previously dedicated to height, to reproduction (Specht & Williams, 1984). Despite this, it is rare to observe similar correlations between increased yield and smaller plant size in wild plants. It is possible that agricultural varieties are unique in displaying this relationship or it may be that the relationship between plant size and reproductive output is more complex than previously assumed by plant ecologists.

The localized over-proliferation of roots in response to interplant root competition was robust to a number of potentially confounding factors. Split-root seedlings were trimmed to have two or three equal-length roots. One might expect the starting number of roots in a pot to influence the final root proliferation, yet the plants achieved a whole-plant allocation strategy and produced the same final root mass in a pot whether they started with one or two roots in the pot. This occurred despite the fact that the plants maintained all of the

roots they started with and did not allocate all new growth to just one root when starting with two roots in the same pot.

The root over-proliferation pattern was also robust to the various space geometries and plant positions in the two three-pot experiments, suggesting that pot geometry, encountering nutrient depletion zones, and initial direct root contact are unlikely to be the cause of the root allocations observed in these experiments. Although we still do not know the precise mechanisms for these responses, there are several studies suggesting that plants are capable of distinguishing their roots from those of neighbors. Chemicals produced by *Larrea tridentata* have been shown to slow the relative growth rate of the roots of neighbors, and physical contact appears to have similar consequences for *A. dumosa* (Mahall & Callaway, 1991, 1992, 1996). The same peas we used in these experiments have also been shown to be able to discriminate between their own roots and the roots of others (Falik *et al.*, 2003). Peas reduce root growth in the direction of their own roots and may even reduce root growth in the direction of genetically identical plants (Falik *et al.*, 2003). Our results agree with the findings of these studies.

Without altering the general root allocation pattern of plants, both nutrient amount and supply rate had significant, albeit different, effects on root production. The significant interaction of nutrient amount with the competition treatments suggests that competition was more intense for peas at low nutrient levels. Among pairs of plants that shared two pots, 'fence-sitters' produced from 7% (0.6 nutrient concentration) to 51% (0.1 nutrient concentration) more roots than those individuals that exclusively occupied single pots as 'owners'. Nutrient supply rate significantly altered total plant biomass such that, in the single pulse treatments, total plant biomass was *c.* 32% higher than when the same amount of nutrients was spread over five waterings (although it did not alter the general phenomenon of root over-proliferation). This is the opposite result to that obtained by Novoplansky & Goldberg (2001) in a pulsed-water experiment. The perennial grasses they studied produced more biomass when pulses were smaller and more frequent. The differences between water and nutrient movement through the soil may be the cause of the observed differences.

Treatments with higher total root production had concomitantly higher shoot production. Shoot:root ratios remained relatively constant, in accord with Gersani *et al.* (1998). The comparison of the competition treatments shows us that the change in root allocation is isolated to the shared patch and is not occurring at the level of the entire plant. This suggests that the differences in root and root : shoot allocation, or lack thereof, may be a direct response to neighbors rather than an allometric side effect from a decrease in plant size as observed by Cahill (2003).

Our results seem to be at odds with work showing that plants grow roots away from neighbors. The repelling effect of the roots of a neighbor has been viewed as a mechanism for

curtailing competition between neighbors (Schenk *et al.*, 1999 and references therein). Rather than being at odds, the different results may simply be revealing different facets of root interactions. Given the opportunity, a plant favors proliferating roots in unexploited rather than exploited soil. In most experiments where roots have been shown to grow away from a the roots of a neighbor, the types of interactions have differed significantly from those studied here (Brisson & Reynolds, 1994; Gersani *et al.*, 1998). In our model, over-proliferation is with respect to the option of investing root growth in space already occupied by the roots of the individual vs space occupied by the roots of a neighbor when the interaction is symmetric and is not influenced by allelopathy (Gersani *et al.*, 2001).

The two-pot experiments of Gersani *et al.* (1998) provide an interesting comparison to our three-pot experiments. They examined root proliferation and seed yield in split-root peas that straddled two pots. One pot was the exclusive domain of the plant while the other was occupied by competitor plants that did not occupy any other pots. Their experimental treatment involved varying the number of competitors in the competition pot. As the number of competitors increased, total root biomass in the competition pot increased. In response, the focal plant reduced root proliferation in the competition pot and increased proliferation in the exclusive pot. The key difference between this study and theirs is that the plants in this study engaged in symmetric interactions. That is, the plants were equal competitors with no advantage or disadvantage over the other. The plants in Gersani *et al.* (1998) were engaged in highly asymmetric interactions where the competitors added did not have the same options as the focal individuals. This asymmetry could result in the different root growth strategies observed for the focal individual and the competing neighbors compared with plants in symmetric interactions. Interestingly, adding more competitors into the competition pot did not significantly decrease the seed yield of the focal plant, although the trend was for lower yield with increased competitor numbers. As in our three-pot experiment, having an exclusive pot may allow a plant to greatly ameliorate the negative effects of root competition elsewhere.

A test of the predictions of the model with wild plants is a next important step. If root over-proliferation and the concomitant decrease in reproductive output are common in nature, this may help to explain nonallelopathic root segregation (Schenk *et al.*, 1999). Humans are able to avoid overgrazing public lands via legislation. The segregation of roots in the absence of harmful chemicals may be the result of plant populations evolving a form of legislation given the obvious benefits of restraining root proliferation in potentially shared spaces. Populations of *A. dumosa* only display segregation with plants from their own population (Mahall & Callaway, 1996). They also show segregation with clones (Mahall & Callaway, 1996), which may imply a role for kin selection in promoting cooperative segregation. A recent study by Bais

et al. (2003) showed plants interacting directly via allelopathic chemicals. It seems possible that in the near future we will also see indisputable direct evidence for other types of direct chemical and physical interactions between plants that may explain the mechanisms behind our results and those of others.

Whatever the mechanism turns out to be, plants appear capable of making game-theoretic choices regarding root growth into unoccupied and shared space. An individual that over-proliferates roots in a population of plants that do not do likewise wins a greater proportion of the available resource pool. An individual that over-proliferates in a population of plants that display the same strategy avoids the 'sucker' payoff that would occur if it were the only individual to restrain root proliferation in the shared spaces. The results presented here show that plants are capable of altering their root growth strategies at scales smaller than their entire root system, and that this may ameliorate the loss in reproductive output observed when plants only have access to areas that they must share with other plants. Thus the negative correlation between root growth and yield, as observed by Gersani *et al.* (2001), may not occur in all contexts. However, there is still much that we do not know about the variables that influence the root growth strategy of a plant and the consequences for plant fitness and this should be an area of fruitful research for some time to come.

Acknowledgements

We thank T. Caravello, N. Gersani, S. Green, J. Moll, P. Reides and Lawrence Sykora for invaluable assistance in cultivating the plants and collecting data. B. Mahall, A. Fitter, J. Cahill, W. Jackson, B. Kotler, M. Rosenzweig, C. Whelan, the Brown Laboratory and three anonymous reviewers provided helpful comments and insights on the ideas and manuscript. The research was supported by a Doctoral Improvement Grant from NSF (E.E.O. and J.S.B.) and by NSF grants (Henry F. Howe and J.S.B.). This work is in partial fulfillment of the requirements for a PhD at the University of Illinois at Chicago.

References

- Bais HP, Park S, Weir TL, Callaway RM, Vivanco JM. 2004. How plants communicate using the underground information superhighway. *Trends in Ecology and Evolution* 9: 26–32.
- Bais HP, Vepachedu R, Gilroy S, Callaway RM, Vivanco JM. 2003. Allelopathy and exotic plant invasion: From molecules and genes to species interactions. *Science* 301: 1377–1380.
- Brisson J, Reynolds JF. 1994. The effect of neighbors on root distribution in a creosotebush (*Larrea tridentata*) population. *Ecology* 75: 1693–1702.
- Cahill JC. 2003. Lack of relationship between below-ground competition and allocation to roots in 10 grassland species. *Journal of Ecology* 91: 532–540.
- Cahill JC, Casper BB. 2000. Investigating the relationship between neighbor root biomass and belowground competition: field evidence for symmetric competition belowground. *Oikos* 90: 311–320.
- Day KJ, John EA, Hutchings MJ. 2003. The effects of spatially heterogeneous nutrient supply on yield, intensity of competition and root placement patterns in *Briza media* and *Festuca ovina*. *Functional Ecology* 17: 454–463.
- Falik O, Reides P, Gersani M, Novoplansky A. 2003. Self/non-self discrimination in roots. *Journal of Ecology* 91: 525–531.
- Gersani M, Abramsky Z, Falik O. 1998. Density-dependent habitat selection in plants. *Evolutionary Ecology* 12: 223–234.
- Gersani M, Brown JS, O'Brien EE, Maina GM, Abramsky Z. 2001. Tragedy of the commons as a result of root competition. *Journal of Ecology* 89: 660–669.
- Gleeson SK, Fry JE. 1997. Root proliferation and marginal patch value. *Oikos* 79: 387–393.
- Hammerstein P. 1998. What is evolutionary game theory?. In: Dugatkin LA, ed. *Game theory and animal behavior*. New York, USA: Oxford University Press, 3–15.
- Hardin G. 1968. The tragedy of the commons. *Science* 162: 1243–1248.
- Hardin G. 1998. Extensions of 'the tragedy of the commons'. *Science* 289: 682–683.
- Hoagland DR, Arnon DI. 1950. *The water culture method for growing plants without soil*. Berkeley, CA, USA: California Agricultural Experiment Station, Circular no. 374.
- Holzappel C, Alpert P. 2003. Root cooperation in a clonal plant: connected strawberries segregate roots. *Oecologia* 134: 72–77.
- Inderjit, Weston LA. 2002. Root exudates: an overview. In: De Kroon H, ed. *Root ecology*. Heidelberg, Germany: Springer-Verlag, 235–255.
- King DA. 1993. A model analysis of the influence of root and foliage allocation on forest production and competition between trees. *Tree Physiology* 12: 119–135.
- Mahall BE, Callaway RM. 1991. Root communication among desert shrubs. *Proceedings of the National Academy of Sciences, USA* 88: 874–876.
- Mahall BE, Callaway RM. 1992. Root communication mechanisms and intracommunity distributions of two Mojave Desert shrubs. *Ecology* 73: 2145–2151.
- Mahall BE, Callaway RM. 1996. Effects of regional origin and genotype on intraspecific root communication in the desert shrub *Ambrosia dumosa* (Asteraceae). *American Journal of Botany* 83: 93–98.
- Maina GM, Brown JS, Gersani M. 2002. Intra-plant versus inter-plant root competition in beans: Avoidance, resource matching or tragedy of the commons? *Journal of Plant Ecology* 160: 235–247.
- Novoplansky A, Goldberg DE. 2001. Effects of water pulsing on individual performance and competitive hierarchies in plants. *Journal of Vegetative Science* 12: 199–208.
- Poehlman JM. 1986. *Breeding field crops*, 3rd edn. New York, USA: Van Nostrand Reinhold.
- Schenk HJ, Callaway RM, Mahall BE. 1999. Spatial root segregation: Are plants territorial? *Advances in Ecological Research* 28: 145–180.
- Schieving F, Poorter H. 1999. Carbon gain in a multispecies canopy: the role of specific leaf area and photosynthetic nitrogen-use efficiency in the tragedy of the commons. *New Phytologist* 143: 201–211.
- Specht JE, Williams JH. 1984. Contribution of genetic technology to soybean productivity – retrospect and prospect. In: Fehr WR, ed. *Genetic contribution to yield gains of five major crop plants*. CSSA Special Publishers 7. Madison, WI, USA: Crop Science Society of America and American Society of Agronomy, 49–74.
- Volis S, Shani U. 2000. The effect of neighbors on the root system of the desert annual *Eremobium aegyptiacum*. *Folia Geobotanica* 35: 161–168.