

The effect of initial seed density on the structure of a desert annual plant community

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Summary

1 Few experiments directly test the role of density dependence in natural plant communities.

2 We tested (i) whether different initial seed densities of the entire seed bank of an annual plant community affected performance (emergence, mean plant biomass and survival) and (ii) whether density-dependent processes were operating within the communities. We also tested whether life-stage, year and soil moisture (both on a topographic gradient and in an experimental manipulation) influenced the effects of seed density.

3 We considered two distinct phases: seed to emergent seedlings, and emergent seedlings to established plants.

4 The seed bank was collected from a semistabilized sand dune in the Negev Desert, Israel. This was added to sieved sand in plots at the same site at four different initial seed densities: 1/16 \times , 1/4 \times , 1 \times (natural seed density) and 2 \times . The experiment was repeated for three consecutive growing seasons.

5 Emergence of seedlings was significantly influenced by initial seed density in all 3 years, with higher initial seed densities having lower rates of emergence.

6 Mean final plant size was negatively density dependent and consistently unaffected by the initial seed density sown.

7 In general, there were no strong interactions of topographic position with initial seed density, processes within the vegetation were not density dependent and the experimental addition of water did not influence any of the performance measures tested. Density-dependent processes vary from year to year, while moisture effects do not.

8 We conclude that seedling emergence and some processes in the established plant community are density dependent, but the established plant community is also affected by other processes, such as resource limitation.

Key-words: annual plants, competition, density dependence, life-stage, soil moisture, topographic position.

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Introduction

Whilst Crawley (1997) argues that populations without density-dependent processes will go extinct, White (2001) denies the very existence of density-dependent regulation. Perhaps the debate was foreseen by Murray (1994), who suggested that although we make general predictions using the concept of density dependence, we do not address specific questions about how regulation might occur. In addition, Goldberg *et al.* (2001)

specifically identify three major shortcomings in the density-dependence literature: (i) most studies only consider variations in density of a single focal species; (ii) most studies that address density dependence in the field only do so non-experimentally; and (iii) observing changes in the density of only a few species in the field makes it difficult to extrapolate to other species or habitats. However, because different species of plants interact in the field, the whole community may be subject to density-dependent regulation. Community-level processes may thus regulate parameters such as birth or death rates and thereby influence community structure (Crawley 1997); however, this general concept and the evidence itself are equivocal (Goldberg & Barton 1992; Murray 1994; Goldberg *et al.* 2001; White 2001). It is

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critical that experimental tests of important ecological concepts, such as density dependence, be validated under more natural conditions.

We are interested in determining whether whole community manipulations of density in the field, through experimental manipulation of the seed bank, directly influence interactions within the plant community, as a direct extension of the study of Goldberg *et al.* (2001), which tested for density dependence under the controlled conditions of an experimental garden. We used the same experimental approach changing initial seed density, but because we also incorporated a topographic gradient (which corresponds to natural differences in soil moisture) and experimental manipulation of water levels, we were able to test whether such regulation or environmental limitation is more important in structuring this plant community.

The potential range of responses by a community to manipulation of total density include negative density dependence (i.e. decreased performance with increasing density, Goldberg & Barton 1992; Goldberg *et al.* 2001), positive density dependence (Bertness & Callaway 1994; Callaway 1995; Callaway & Walker 1997; Goldberg *et al.* 2001) and density independence. Negative density dependence is typically interpreted as either competition through interference or exploitation, and positive density dependence is typically attributed to facilitative non-trophic interactions such as reduced stress or protection from herbivores (Bertness & Callaway 1994; Callaway 1995, 1998). The most direct methods used to test for these effects are removal of the entire neighbourhood surrounding a focal species and sowing different densities of the community. The first approach is common in both the competition and facilitation literature (Goldberg & Barton 1992; Callaway 1995). While the second is rarer (Goldberg *et al.* 2001), it provides the opportunity to test for two different effects. First, it determines whether seed–seed interactions may influence the early emergent and final established plant community, and secondly, it provides a test of density dependence without altering the proportions of species represented within both the germinating and emergent vegetation. Competition studies that measure net effects at the end of the growing season have been criticized for recording only the final outcome of interactions (Gibson *et al.* 1999; Connolly *et al.* 2001), because it is highly likely that the outcome of interactions between plants is dependent on life-stage, primarily due to changes in plant size (Callaway & Walker 1997). The manipulation of initial community density (Goldberg *et al.* 1995; Goldberg *et al.* 2001) and monitoring changes in the subsequent plant community over time allows us to distinguish between interactions and final outcomes (i.e. density-dependent processes and density-dependent effects). By changing the independent variable for different dependent variables based on life-stage, i.e. seed density for emergence and final plant density for shoot mass, Goldberg *et al.* (2001) implicitly assume that the initial seed density

treatments are important only for initial early season measures such as emergence. However, this may not always necessarily be the case. We explicitly tested whether interactions between seeds are important for both the germinating plant community and the emergent plant community by using two separate sets of analyses: ANOVAs for response variables to the initial seed density manipulations (additional approach) and regressions for interactions within each life-stage (similar to Goldberg *et al.* 2001). To test for density dependence, the latter approach is sufficient, but both approaches are necessary to fully understand the relative importance of interactions between seeds and plants and to distinguish between interactions and outcomes.

In addition to changes in density-dependent effects with life-stage, the level of environmental stress may also influence the outcome of interactions between plants, which is inversely related to productivity. The predictions are highly debated (Goldberg & Barton 1992; Goldberg *et al.* 1999), but in general competition is either expected to decrease with increasing stress (Grime 1973, 1977; Bertness & Callaway 1994) or remain relatively constant (Newman 1973; Tilman 1988). It has also been argued that the type of stress gradient tested (natural versus manipulative) may significantly influence whether the outcome is positive or negative (Goldberg & Barton 1992; Kadmon 1995). Furthermore, the level of stress, in these cases available water, may also vary from year to year depending on local weather conditions such as total amount or frequency of rainfall (Kadmon 1995; Tielborger & Kadmon 2000; Novoplansky & Goldberg 2001). In this study, we restrict our treatment of stress to moisture level by using two different topographic positions on a dune that indirectly corresponds to a natural soil moisture gradient (high and low soil moisture) and by supplementing natural precipitation in the field at both positions. We manipulate both moisture level and density concurrently to determine whether the importance of available water at different seed and plant densities varies and if limitation by the environment is important.

Using the initial community density approach (developed by Goldberg *et al.* 1995), i.e. sowing the seed bank at different densities, we ask the following questions: (i) does the initial seed density influence the emergent and the established plant community; and (ii) are density-dependent processes important within both of these communities. We predicted that density effects would be observed, whereby initial seed density influences the plant community both at the emergent seedling stage and in its final form, and interactions within the established plant community will also be density dependent. We also hypothesized that the effect of initial seed density and density-dependent effects will vary with life-stage, moisture (i.e. with topographic position on a natural soil moisture gradient and manipulation of amount of water) and temporally (i.e. between years).

Methods

STUDY SITE AND ANNUAL PLANT COMMUNITY

The study site is a semistabilized sand dune dominated by winter annuals and sparsely distributed shrubs at Bir Asluj in the Holot Mashabim Nature Reserve (31 00.212' N, 34 44.474' E) in the central Negev desert, Israel. There are typically about 20 species of annual plants in this community (area of dune about 1 ha) (Dyer *et al.* 2001; Goldberg *et al.* 2001). Rainfall occurs primarily in winter months (December to April) and has a 30-year average of 110 mm per year (Goldberg *et al.* 2001). In the 1998 growing season there were 27 rainfall events totalling 96.82 mm; in 1999, rainfall events were less frequent (10 totalling 39.6 mm), and in 2000, 14 rainfall events totalled 35.7 mm (data provided by the Blaustein Meteorological Unit). Hence, the years sampled included one relatively wet year and two dry years (1/3 the MAP). The experiment was performed on the north face of the dune with prevailing winds from the north-west. Soil moisture was measured five times each season with a portable TDR (Tektronix 1502C) at two positions on the dune (lower and upper) and at two depths (5 and 20 cm, field capacity: 16% soil moisture). The lower position on the dune consistently had higher soil moisture at both depths (mean percentage field capacity; $11.6 \pm 0.07\%$ vs. $10 \pm 0.08\%$ at 5 cm, $47.3 \pm 0.67\%$ vs. $44.3 \pm 0.7\%$ at 20 cm, $F_{3,108} = 4.78$, $P = 0.0035$, and $F = 6.94$, $P = 0.0003$, respectively, repeated measure ANOVAS). The growth of natural vegetation and transplants also confirms that the upper position on the dune is less suitable for plant growth (lower productivity) than the lower position.

EXPERIMENTAL DESIGN

To test for density dependence, initial seed density was varied by sowing different amounts of the entire seed bank (thereby maintaining a constant proportion of the different species of seeds into 0.5×0.5 m plots). The experiment was repeated during the winters of 1998, 1999 and 2000 at the two different topographic positions on the dune (corresponding to high and low soil moisture). In 1998 the species abundance was recorded throughout the growing season to determine if seed density affects diversity. In 1999, half of the plots at each position received additional water to further test if moisture level influenced density-dependent processes.

Seed was collected (along the length of the dune) early each November using a modified shovel that removes the top 2 cm of sand. This depth effectively samples at least 99% of the total seed bank (Venable 1989). Seed was separated from the sand using a 500- μ m sieve to capture virtually all of the seed (Goldberg *et al.* 2001). Natural seed bank density (1 \times) was then

calculated for the field site (based on amount of seed collected per unit area) and was found to be 600.8 g m^{-2} in 1998, 625.3 g m^{-2} in 1999 and 597 g m^{-2} in 2000 (mean = 607.7 g m^{-2}). Plots were sown at one of four seed densities: 1/16 \times , 1/4 \times , 1 \times and 2 \times the natural seed density for the field (1998 value used for all three seasons).

Two transects (50 m apart) were marked, one at the lower position (higher soil moisture) and one at the upper position (lower soil moisture) on the same semistabilized sand dune from which seed had been collected. On each transect, 10 blocks each with 5 plots (0.5×0.5 m) were marked. The blocks were spaced at least 1 m apart on the transects, and plots had a 20-cm buffer between them. The sand from four of the plots in each block was collected and sieved to remove seed before mixing with collected seed to give one of the four densities and replaced in each plot. Densities were randomly assigned to plots within each block. In 1998, 10 additional 0.5×0.5 m plots were cleared, sieved, and the seedless sand replaced to test the effectiveness of sieving in the field and to determine if additional seeds were dispersed into plots prior to germination.

Each year the emergent vegetation was surveyed three times: early season (the end of January after the maximum number of seedlings had emerged as determined by two to three initial censuses), mid-season (the end of February), and at the end of the growing season (the second to third week of April). Abundance of each species was recorded in 1998 and total plant density was recorded in 1999 and 2000 for each census. At the end of the growing season all above-ground vegetation was harvested. The plants were dried at 60 °C for 48 h and weighed. In 1998, all sand (top 2 cm) and remaining material were also collected from the plots (and sieved) to provide an estimate of seed production per plot.

In 1999, half of the blocks at both positions on the dune were randomly assigned to an additional water treatment. Control blocks received only natural rainfall and treatment blocks were hand watered with a very fine nozzle early in the morning receiving 1 L of water per plot every 10 days (equivalent to 100 mm of precipitation in addition to rainfall).

ANALYSES

The effect of seed density on the plant community was primarily tested by comparing the emergence index, proportion surviving and mean final plant size. The emergence index is a measure of the probability of emerging and was calculated by dividing the total number of emergent seedlings by the number of seeds added. The mass of seed added to each plot was converted to seed number by dividing by the calculated mean number of seeds per gram (based on trial counts and average seed sizes). The proportion surviving (from the total number of emergent seedlings) was calculated for the mid-season and end of season censuses,

and mean plant size (biomass/density) was calculated at the end of the growing season. Differences between groups for each of the three response variables were tested by repeated measures ANOVAs (for proportion surviving throughout the season) or ANOVAs (emergence index and mean final plant size) using JMP 4 (SAS 2000). The main effects were initial seed density, topographic position (lower-upper), census (for proportion surviving) and their interactions. In 1998, species diversity, E_{var} (Smith & Wilson 1996), was also tested as a response variable. The diversity data were also compared with a null model of increasing diversity with increased seed density due to a sampling effect (Goldberg & Estabrook 1998). In 1999, supplemental water addition was also tested as a main effect in the ANOVA models.

Density dependence within the plant community at different life-stages and topographic positions was tested each year using regression analyses similar to Goldberg *et al.* (2001), including linear, power ($\ln(x)$ and $\ln(y)$), semilog ($\ln(x)$ (y), and reciprocal (x , $1/y$) functions. The model of best fit (highest R^2) is reported. The emergence index was regressed against initial seed density, and mean plant size and final proportion surviving were regressed against final plant density. It was predicted that the population level response (final proportion surviving) should increase linearly with the number of seeds planted if there is no density dependence in the community (null). Non-linearity indicates density dependence (concave = competition and convex = facilitation). For the individual performance measures, the emergence index and mean plant size, the null relationships have zero slopes, i.e. both the probability of an indi-

vidual seed emerging and plant size are independent of density. A significant positive slope suggests facilitation and a negative slope competition.

Differences between years in the effects of initial seed density on all performance measures were also compared using ANOVAs. The main effects were year, topographic position, seed density and interactions with year. Only comparable plots were included in these analyses, i.e. plots in 1999 that received additional water were not included.

Results

CONTROL PLOTS

The control plots tested in 1998 had a small number of seeds germinate (mean = eight plants), but these numbers were significantly less than the emergent plant density of all other plots (ANOVA, $F_{5,72} = 13.73$, $P = 0.0001$, with contrast analyses $P < 0.001$, mean for lowest emergent plant density from a treatment plot, $1/16 \times = 50.3$ plants).

DENSITY EFFECTS

Initial seed density effects

The analyses of initial seed density effects within each year and between years demonstrated a range of responses from no effect of low initial seed density to significant negative effects at higher seed densities. In all 3 years, the emergence index generally decreased with higher seed densities (Fig. 1a, Table 1), whereas the proportion surviving throughout the

Table 1 ANOVAs for final response variables to initial seed density manipulations in 1998, 1999, 2000 (df_{denominator} = 71). Sequential Bonferroni corrections were applied for each response variable and only significant values are in bold. Differences in proportion surviving were tested with a full-factorial repeated measures ANOVA as proportion surviving to each census was calculated. There were no significant interaction effects with census so only the main effect is reported here. Position refers to topographic position on the dune (lower and upper, which correspond to high and low soil moisture, respectively), and water to the addition of water to half the blocks (1999 only)

Measure	Effect	d.f.	1998			1999			2000		
			SS	F ratio	P-value	SS	F ratio	P-value	SS	F ratio	P-value
Emergence index	Position	1	0.0000	8.1	0.012	0.0007	1.5	0.23	0.0003	15.9	0.0003
	Seed density	3	0.0004	54.6	0.0001	0.05	37.1	0.0001	0.0008	12.43	0.0001
	Pos. × SD	3	0.0000	4.3	0.021	0.0001	0.49	0.69	0.0005	7.6	0.0004
	Water	1				0.0001	0.14	0.71			
Proportion surviving	Position	1	0.001	0.03	0.86	0.0000	0.0001	0.99	2.24	38.9	0.0001
	Seed density	3	0.15	1.6	0.21	1.3	6.23	0.0005	0.6	3.43	0.021
	Pos. × SD	3	0.09	0.96	0.42	1.5	7.2	0.0001	0.69	4.0	0.011
	Census	1	0.53	17.4	0.0002	15.1	109.0	0.0001	1.63	28.45	0.0001
	Water	1				0.06	0.83	0.36			
Mean plant size	Position	1	0.32	6.43	0.022	0.22	0.93	0.34	0.008	3.5	0.07
	Seed density	3	0.43	3.0	0.064	0.71	1.0	0.40	0.014	1.94	0.14
	Pos. × SD	3	0.16	1.1	0.39	0.7	0.99	0.41	0.003	0.47	0.7
	Water	1				0.24	1.02	0.32			

SD = seed density.

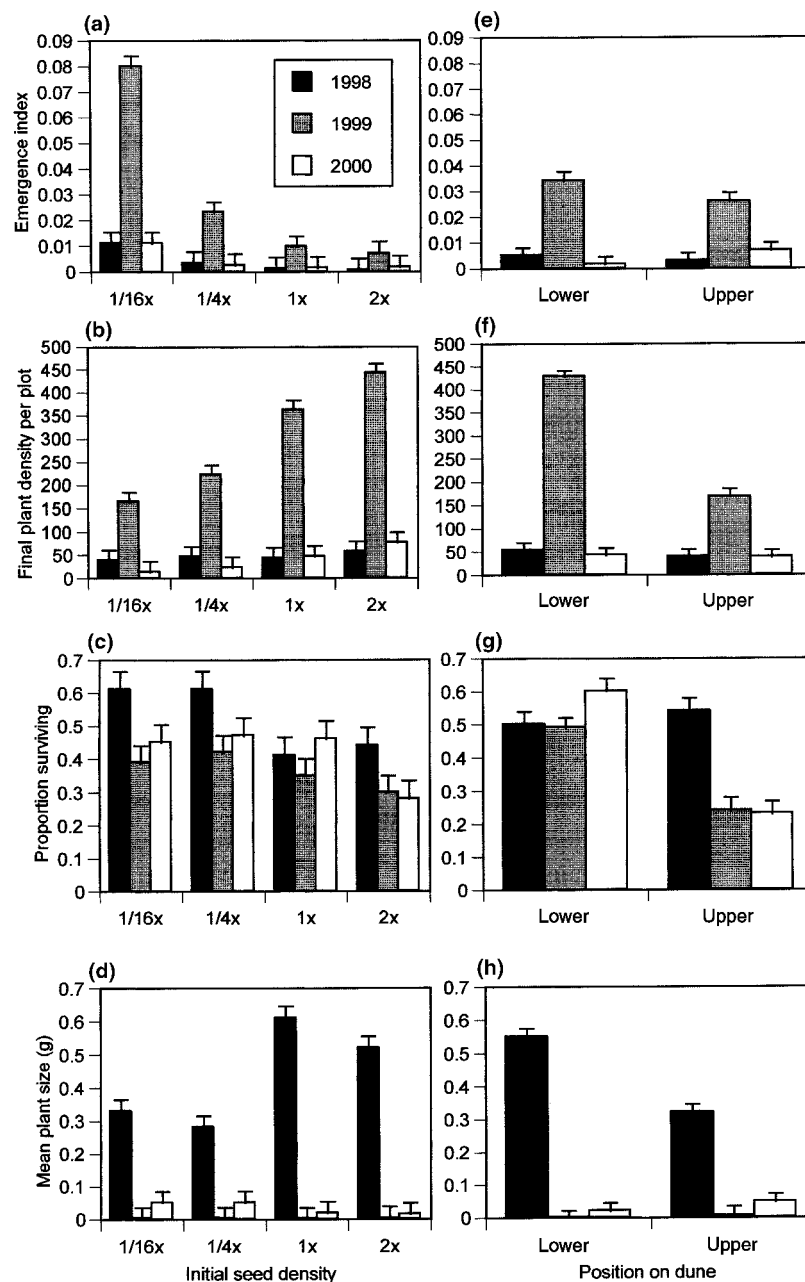


Fig. 1 A comparison of effects of initial seed density (relative to natural seed density in the field) and topographic position on the performance of an annual plant community. Seeds were sown prior to the growing season (November) at lower and upper sites (high and low soil moisture, respectively). The emergence index is the proportion of seeds that germinated based on total (maximum) emergence. The proportion surviving is the number of individuals at each census/total emergence density (from repeated measures ANOVAS with no significant interactions between census and seed density, $P > 0.05$). Mean plant size was calculated as total biomass per plot/total final plant density. The error bars are \pm SE.

season and mean plant size were unaffected (Fig. 1c,d, Table 1). There were some differences, but they were not consistent across years and the correction for multiple tests made most trends statistically non-significant (Table 1). Plant diversity was also unaffected by initial seed density (ANOVAS, $P > 0.25$). Although species diversity increased in higher seed densities (data not shown), observed diversity was frequently much lower than the null expected at a given density ($P > 0.05$, same approach as Goldberg & Estabrook 1998).

Density dependence within the plant community

Regression analyses detected both density-dependent and independent processes. In 1998, only the emergence index demonstrated density dependence (Fig. 2a, Table 2); the relationship was negative (slope of line significantly different from 0 and negative) and shows competition at higher densities. There were no differences between the two positions on the natural soil moisture gradient for any response variable in 1998 (ANOVAS, $P > 0.05$). The topographic positions did not

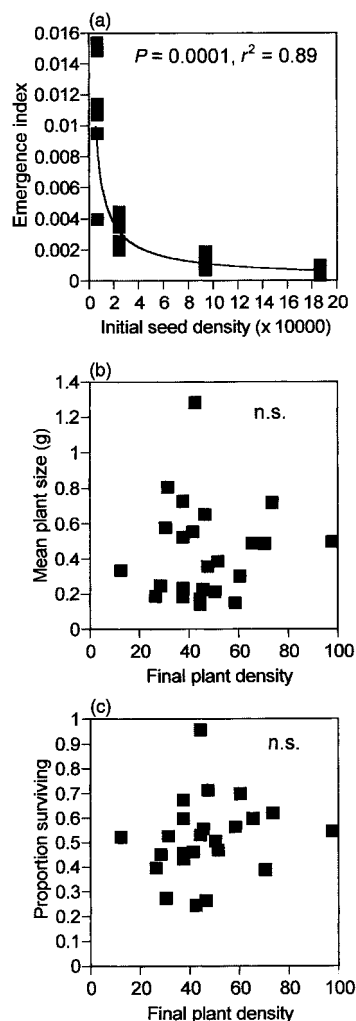


Fig. 2 The effects of density on the performance of an annual plant community in the 1998 growing season. High and low sites are combined. Regression coefficients and fitted models are reported in Table 2.

Table 2 Regression coefficients for interactions within the emergent vegetation. Position refers to topographic position on the sand dune. In 1998, the two sites were pooled as there were no significant differences between them for any of the response variables (ANOVAS, $P > 0.05$). The P -values reported here refer to whether the slope of the line is significantly different from zero. Significant P -values are in bold after sequential Bonferroni corrections. The degrees of freedom reported are for the total model (model + error), and the model refers to the set of transformations that best fit the data (see text for description of transformations for each model)

Year	Position	Measure	Model	d.f.	Intercept	Slope	R^2	P -value
1998	Pooled	Emergence index	Power	23	2.2	-0.79	0.89	0.0001
		Plant size	Linear	23	0.38	0.001	0.005	0.73
		Prop. surviving	Linear	23	0.44	0.002	0.04	0.37
		Seed production	Linear	22	0.0005	-5×10^{-6}	0.002	0.85
1999	Lower	Emergence index	Power	39	2.4	-0.59	0.79	0.0001
		Plant size	Linear	75	0.002	-3×10^{-6}	0.41	0.0001
		Prop. surviving	Power	39	-1.82	0.16	0.12	0.0253
1999	Upper	Emergence index	Power	39	2.43	-0.62	0.77	0.0001
		Plant size	Linear	79	0.016	-0.0001	0.44	0.0001
		Prop. surviving	Reciprocal	38	5.51	-0.006	0.12	0.03
2000	Lower	Emergence index	Semilog	22	0.009	-0.00073	0.20	0.033
		Plant size	Power	18	-1.94	-0.63	0.42	0.0026
		Prop. surviving	Linear	23	0.58	0.00043	0.003	0.8
2000	Upper	Emergence index	Semilog	23	0.06	-0.0047	0.48	0.0002
		Plant size	Semilog	23	0.19	-0.046	0.61	0.0001
		Prop. surviving	Linear	22	0.16	0.0009	0.08	0.18

differ in 1999; both the emergence index and mean plant size were negatively density dependent (Fig. 3a-d, Table 2), while the proportion of individuals surviving was density independent (Fig. 3e,f, Table 2). In 2000, the emergence index was negatively density dependent only for the upper position on the dune but mean plant size was negatively density dependent for the lower position (Fig. 4a-d, Table 2). Proportion surviving was density independent at both positions (Fig. 4e,f). Overall, the emergence index and mean plant size were generally negatively related to density, while proportion surviving was density independent.

LIFE STAGE

Earlier life stages were generally more affected by initial seed density (i.e. negative effects on seedling emergence, Fig. 1a) than later ones (final mean plant size unaffected by initial seed density in all 3 years, Fig. 1d). Density-dependent processes within the plant community were generally negative early in the growing season (regression of emergence indices on initial seed density) but became either negatively density dependent (mean plant size versus final plant density) or density independent (proportion surviving in all cases). The proportion of individuals surviving within each season significantly decreased with time (Table 1 with contrast analyses for census times, $P < 0.01$).

MOISTURE

The level of natural soil moisture influenced the plant communities by significantly reducing growth at the upper position on the dune (Fig. 1e,f,g,h) but generally did not interact with initial seed density (Table 1). Similarly, density-dependent processes within the

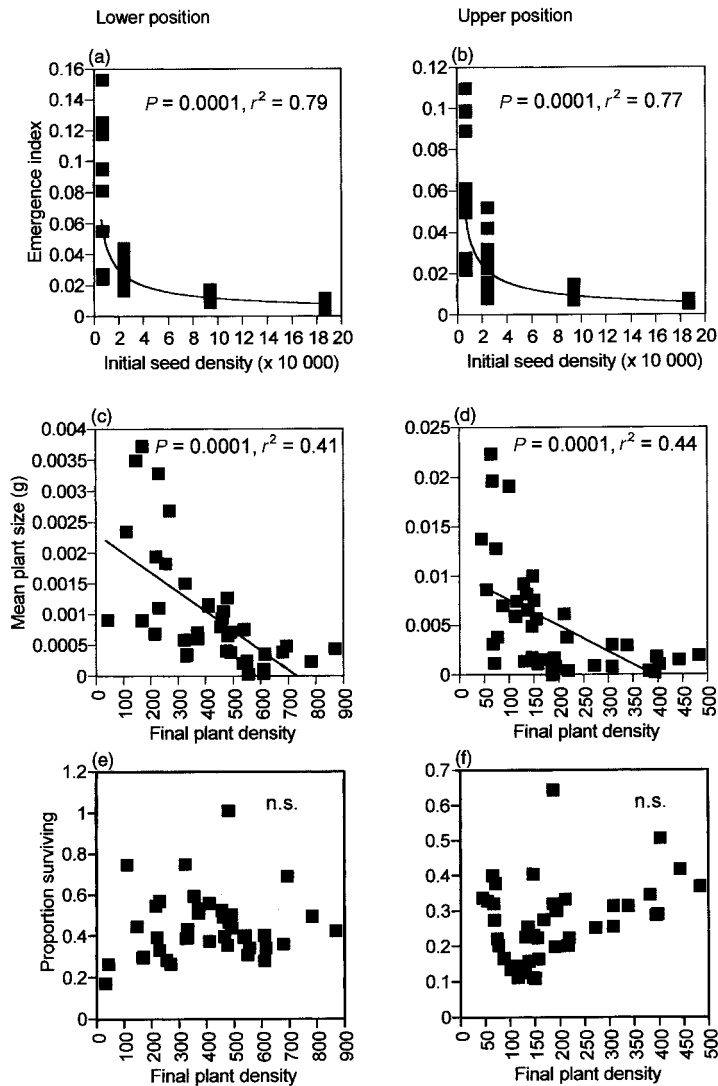


Fig. 3 The effects of density on the performance of an annual plant community in the 1999 growing season. Regression coefficients and fitted models are reported in Table 2.

community generally did not differ with topographic position, except in 2000 when the emergence index was negatively density dependent only at the upper position (Table 2). There was also no effect of water addition in 1999 on any performance measure for initial seed densities or density dependence within the community (Table 1).

TEMPORAL EFFECT

There were significant differences between the 3 years tested in terms of the emergence index, plant density, proportion surviving and mean plant size (Table 3). The highest emergence and final plant density were in 1999 (Fig. 1a,b), and the largest plants and the highest proportion surviving (upper dune only) in 1998 (Fig. 1c,d). Density-dependent processes within the community also changed between years (Figs 2–4). The 2 dry years (1999, 2000) were similar insofar as they expressed differences between positions (in emergence), while the wettest year, 1998, did not.

Discussion

Density-dependent processes are typically tested at the population level. Unfortunately, a shortcoming of this approach is that the target population is typically embedded in a matrix of neighbouring species, i.e. a community. This shortcoming has been addressed by Goldberg *et al.* (2001) by using the initial community density approach (Goldberg *et al.* 1995), which generates a range of population densities for *all* of the species typically represented in the community. However, a second difficulty is that common perceptions about density-dependent processes are not explicitly formulated nor tested, for example: What parameter is being regulated? Are density-dependent responses linear? What range of population densities are affected (Murray 1994)? Thirdly, interactions between species (i.e. effects of one species on another) and the outcome of these interactions (e.g. which species dominates) are not clearly differentiated, and density dependent processes likely also vary throughout the life cycle

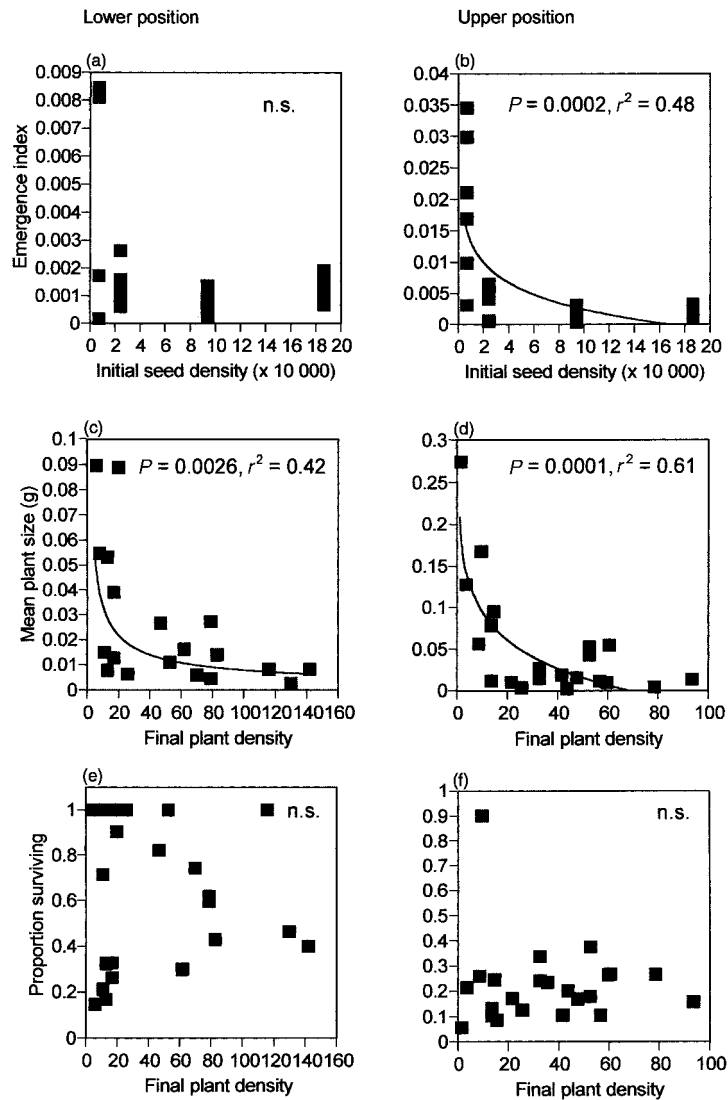


Fig. 4 The effects of density on the performance of an annual plant community in the 2000 growing season. Regression coefficients and fitted models are reported in Table 2.

(Connolly *et al.* 2001). We specifically addressed all three of these difficulties by making specific testable predictions, by measuring interactions within the community and final outcomes, and by differentiating between early season and late season effects by measuring both initial effects of seed density and density-dependent interactions throughout the growing season. This experimental approach thus determines if processes such as emergence or survival have an impact at the community level.

Two questions emerge when we consider a growing plant community. First, do initial starting conditions influence the subsequent structure of the early emergent and later established community? In essence, are initial treatments as important as subsequent interactions on the final plant community? Secondly, what impact do interactions occurring within the developing plant community have on final plant community structure? To address these questions we divided the life cycle of the plants into two distinct phases: seed to emergent seedlings, and emergent seedlings to

established plants. The early phase includes the process of emergence, with the end product being density of seedlings, and the later phase includes survival and growth rate, with the end products being density and dry weight of survivors. The experimental manipulation of seed density generates a range of seed and seedling densities. Increasing seed density decreased the probability of a seedling emerging, while measures of growth or survival at later life stages were unaffected by the initial seed density. Later in the season as the plants grow, interactions between these established plants are more important than the initial seed densities sown (Callaway & Walker 1997; Holmgren *et al.* 1997). However, the community in which these density-dependent processes occur is established by early interactions between seeds and seedling. Analysis of the established plant community showed that there was significant divergence from the emergent plant community. Density-dependent processes at later life stages, chiefly growth rate, determined the final plant community, while initial seed densities did not.

Table 3 ANOVAs for final response variables to initial seed density manipulations across three seasons (1998–2000, $df_{\text{denominator}} = 137$). Interactions between main effects such as position \times seed density are reported in the analyses for each year. Significant *P*-values are in bold (sequential Bonferroni correction did not change table-wide $P < 0.05$ significance)

Measure	Effect	d.f.	SS	<i>F</i> ratio	<i>P</i> -value
Emergence index	Year	2	0.02	47.9	0.0001
	Position	1	0.0001	0.41	0.52
	Seed density	3	0.24	34.1	0.0001
	Year \times position	2	0.001	2.42	0.09
	Year \times seed density	6	0.023	16.6	0.0001
Final plant density	Year	2	2 158 248	180.2	0.0001
	Position	1	317 776	53.1	0.0001
	Seed density	3	320 235	17.82	0.0001
	Year \times position	2	318 451	8.86	0.0001
	Year \times seed density	6	521 787	43.6	0.0001
Proportion surviving	Year	2	0.59	8.38	0.0004
	Position	1	1.4	39.73	0.0001
	Seed density	3	0.59	5.55	0.0013
	Year \times position	2	0.23	1.1	0.38
	Year \times seed density	6	1.1	15	0.0001
Mean plant size	Year	2	5.64	195.4	0.0001
	Position	1	0.16	11.11	0.0011
	Seed density	3	0.24	5.5	0.0014
	Year \times position	2	0.68	7.8	0.0001
	Year \times seed density	6	0.48	16.8	0.0001

The initial effects of seed density and subsequent interaction between plants may not always act in the same direction. For instance, increased seed density strongly reduced the proportion of seeds that emerged in all three seasons, while subsequent survival was not related to increases in plant densities. Depending on the strength of the interactions at different life stages, we may see net positive or net negative outcomes for the final plant community. Admittedly, very few studies have tested for, and detected, interactions between seeds or seedlings. Nonetheless, there are a growing number of studies that have identified both positive (Linhart 1976; Waite & Hutchings 1978; Smith 1983; Adler *et al.* 1993) and negative (Palmbad 1968; Murray 1998; Goldberg *et al.* 2001) effects of manipulating seed density on germination and emergence. It has been shown that seeds may 'sense' other seeds, e.g. by the release of leachates (Murray 1998), and change patterns in germination either through accelerated emergence (Dyer *et al.* 2000) or by reduced emergence (Murray 1998; Goldberg *et al.* 2001). Although we can only speculate, the negative effects of increased seed density may also be a product of chemical inhibition by some of the species of seeds within the community (Qadir & Abbasi 1971). Therefore, it is crucial to measure *both* initial effects of density and subsequent interactions between plants to understand what processes influenced the emergent plant community.

The second question we address was the overall importance of density dependence within the establishing plant community. It appears that competition influences this plant community through its effects on differential growth rate rather than on differential survival. This would typically be interpreted as regulation. But is it regulation or limitation? White (2001) would

argue that decreases in mean plant size could be attributed to limitation of a key resource such as water and that competition is a consequence of shortage in a resource, not a cause. This may be true, particularly as mean plant size was density independent in the wet year and negatively density dependent in the two dry years. However, the negative density dependence for emergence of seedlings is not likely to be related to a limitation in water for the following reasons. The experimental addition of water had no effect on seedling emergence, and emergence did not differ between years (one wet year and two dry years). Density-dependent emergence is thus more reasonably attributed to regulation via interference through the production of leachates as discussed earlier. The density independence of survival throughout the growing season similarly suggests that resource limitation is not important in determining survival once the plants are established. Hence, regulation and limitation may both be important in structuring this plant community, with regulation determining establishment and limitation determining growth. White (2001) is correct in stating that we often assume that the number of individuals in a population or community are regulated below the capacity of the environment without testing if limitation is actually determining the numbers instead. Our results suggest that it is perhaps premature to conclude that there is no regulation at the community level, but that we do need to be cautious when interpreting single measures as *either* limitation *or* regulation.

As expected, the effects of initial seed density and density dependence varied with life stage. The effects of initial seed density were important early in the growing season but had little effect thereafter. It has been proposed that facilitation is more important for earlier life

stages and that as plants increase in size they are more likely to compete (Callaway & Walker 1997). Our results do not support this prediction, with negative density-dependent emergence occurring in all three growing seasons and decreases in mean final plant size with increasing plant density detected in 1999 and 2000. Hence, life stage strongly influences the net interactions within a plant community, with earlier life stages being negative while later life stages were either negative or density independent. Perhaps the generality of detecting competition versus facilitation in the plant ecology literature has been due in part to measurement at single life stages. Successive measurements provide a viable solution. Further work that incorporates experimental changes in density at different life stages for the same community would also be revealing. For instance, changes in initial seed density coupled with experimental changes in plant density at later life stages would test for differences in the importance of density at each life stage. The experimental manipulations could include not only reductions in density via removal of seedlings or adult plants but the addition of seeds or seedlings to emergent plant communities (see Fowler 1986).

We predicted that topographic position and year would also significantly interact with density effects. Whilst topographic position affected the plant community (lower soil moisture site reduced performance), position did not interact with the effects of initial seed density. Furthermore, the lack of a clear difference between sites in the patterns of density dependence support the hypothesis that the net effect of changing productivity is a relatively constant intensity of competition (Newman 1973; Tilman 1988). The regular addition of water to plots also had no appreciable effect on performance or on the effects of density, which suggests that either competitive intensity is relatively constant or that the pulsing of the resource (i.e. frequency of rainfall and duration between events) is more important than the actual magnitude (Novoplansky & Goldberg 2001). The natural soil moisture gradient we used (i.e. position on dune) encompassed a greater range of abiotic conditions than the experimental addition of water as there were reductions in survival and growth at the naturally lower soil moisture site but no differences with water addition. The type of gradient tested thus has the potential to influence our ability to detect plant interactions (Goldberg & Barton 1992; Kadmon 1995). In this dune system, natural soil moisture affects the plant community but does not directly influence the interactions within the plant community (i.e. density-dependent processes). Further work that tests the effects of timing and frequency of limiting resources will determine whether soil moisture (and subsequent productivity) can affect interactions between plants in this capacity.

Overall, our results show that regulation and limitation both played a role in structuring this dune community, with regulation determining emergence and

limitation determining growth rates. The year-to-year variation and lack of strong consistent effects of moisture gradients suggest that interactions within the plant community may not necessarily change with productivity (at least above ground). These results suggest that it is critical to use successive measurements spanning the entire life stage of plants and that community-level net outcomes may differ from interactions between seeds and seedlings, or within the established plant community.

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References

- Adler, L.S., Wikler, K., Wyndham, F.S., Linder, C.R. & Schmitt, J. (1993) Potential for persistence of genes escaped from canola: germination cues in crop, wild, and crop-wild hybrid *Brassica rapa*. *Functional Ecology*, **7**, 736–745.
- Bertness, M.D. & Callaway, R. (1994) Positive interactions in communities. *Trends in Ecology and Evolution*, **9**, 191–193.
- Callaway, R.M. (1995) Positive interactions among plants. *Botanical Review*, **61**, 306–349.
- Callaway, R.M. (1998) Are positive interactions species-specific? *Oikos*, **82**, 202–207.
- Callaway, R.M. & Walker, L.R. (1997) Competition and facilitation: a synthetic approach to interactions in plant communities. *Ecology*, **78**, 1958–1965.
- Connolly, J., Wayne, P. & Bazzaz, F.A. (2001) Interspecific competition in plants: how well do current methods answer fundamental questions? *American Naturalist*, **157**, 107–125.
- Crawley, M.J. (1997) The structure of plant communities. *Plant Ecology* (ed. M.J. Crawley), pp. 475–531. Blackwell Scientific, Oxford.
- Dyer, A.R., Fenech, A. & Rice, K.J. (2000) Accelerated seedling emergence in interspecific competitive neighbourhoods. *Ecology Letters*, **3**, 523–529.
- Dyer, A.R., Goldberg, D.E., Turkington, R. & Sayre, C. (2001) Effects of growing conditions and source habitat on plant traits and functional group definition. *Functional Ecology*, **15**, 85–95.
- Fowler, N. (1986) Density-dependent population regulation in a Texas grassland. *Ecology*, **67**, 545–554.
- Gibson, D.J., Connolly, J., Hartnett, D.C. & Weidenhamers, J.D. (1999) Designs for greenhouse studies of interactions between plants. *Journal of Ecology*, **87**, 1–16.
- Goldberg, D.E. & Barton, A.M. (1992) Patterns and consequences of interspecific competition in natural communities: field experiments with plants. *American Naturalist*, **139**, 771–801.
- Goldberg, D.E. & Estabrook, G.F. (1998) Separating the effects of number of individuals sampled and competition on species diversity: an experimental and analytic approach. *Journal of Ecology*, **86**, 983–988.
- Goldberg, D.E., Rajaniemi, T., Gurevitch, J. & Stewart-Oaten, A. (1999) Empirical approaches to quantifying interaction intensity: competition and facilitation along productivity gradients. *Ecology*, **80**, 1118–1131.

- Goldberg, D.E., Turkington, R. & Olsvig-Whittaker, L. (1995) Quantifying the community-level consequences of competition. *Folia Geobotanica Phytotax*, **30**, 231–242.
- Goldberg, D.E., Turkington, R., Olsvig-Whittaker, L. & Dyer, A.R. (2001) Density dependence in an annual plant community: variation among life history stages. *Ecological Monographs*, **71**, 423–446.
- Grime, J.P. (1973) Competitive exclusion in herbaceous vegetation. *Nature*, **242**, 344–347.
- Grime, J.P. (1977) Evidence for the existence of three primary strategies in plants and its relevance to ecological and evolutionary theory. *American Naturalist*, **111**, 1169–1194.
- Holmgren, M., Scheffer, M. & Huston, M.A. (1997) The interplay of facilitation and competition in plant communities. *Ecology*, **78**, 1966–1975.
- Kadmon, R. (1995) Plant competition along soil moisture gradients: a field experiment with the desert annual *Stipa capensis*. *Journal of Ecology*, **83**, 253–262.
- Linhart, Y.B. (1976) Density-dependent seed germination strategies in colonizing versus non-colonizing plant species. *Journal of Ecology*, **64**, 375–380.
- Murray, B.R. (1994) On density dependence. *Oikos*, **69**, 520–523.
- Murray, B.R. (1998) Density-dependent germination and the role of seed leachate. *Australian Journal of Ecology*, **23**, 411–418.
- Newman, E.I. (1973) Competition and diversity in herbaceous vegetation. *Nature*, **244**, 310–311.
- Novoplansky, A. & Goldberg, D.E. (2001) Effects of water pulsing on individual performance and competitive hierarchies in plants. *Journal of Vegetation Science*, **12**, 199–208.
- Palmblad, I.G. (1968) Competition in experimental populations of weeds with emphasis on the regulation of population size. *Ecology*, **49**, 26–34.
- Qadir, S.A. & Abbasi, M. (1971) Chemical interaction between seeds of common plants. *Pakistan Journal of Scientific and Industrial Research*, **14**, 211–218.
- SAS Institute (2000) *Jmp IN 4.02*. SAS Institute, Cary, North Carolina.
- Smith, B.H. (1983) Demography of *Floerkea proserpinacoides*, a forest-floor annual. III. Dynamics of seed and seedling populations. *Journal of Ecology*, **71**, 413–425.
- Smith, B. & Wilson, B. (1996) A consumer's guide to evenness index. *Oikos*, **76**, 70–82.
- Tielborger, K. & Kadmon, R. (2000) Temporal environmental variation tips the balance between facilitation and interference in desert plants. *Ecology*, **81**, 1544–1553.
- Tilman, D. (1988) *Plant Strategies and the Dynamics and Structure of Plant Communities*. Princeton University Press, Princeton.
- Venable, D.L. (1989) Modeling the evolutionary ecology of seedbanks. *Ecology of Soil Seedbanks* (eds M.A. Leck, V.T. Parker & R.L. Simpson), pp. 67–90. Academic Press, London.
- Waite, S. & Hutchings, M.J. (1978) The effects of sowing density, salinity and substrate upon the germination of seeds of *Plantago coronopus* L. *New Phytologist*, **81**, 341–348.
- White, T.C.R. (2001) Opposing paradigms: regulation or limitation of populations? *Oikos*, **93**, 148–152.

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