RESEARCH *New Phytol.* (2000), **146**, 471–481

Growth characteristics in *Hordeum spontaneum* populations from different habitats

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Received 30 September 1999; accepted 1 March 2000

SUMMARY

Hordeum spontaneum shows a large genetic variation and occupies a wide range of different habitats. The aim of this study was to quantify variation in growth characteristics of *H. spontaneum* from different sites in Israel and to relate this variation to different environmental conditions. To this end, 84 accessions of 21 populations were grown in a growth chamber in near-optimal conditions and a range of physiological, morphological, allocation-related and chemical characteristics were measured. These parameters included rates of photosynthesis, shoot and root respiration, specific leaf area, biomass allocation and seed mass. Averaged over all traits variation explained by differences between populations was 26%, between accessions 21%, whereas that within accessions was 53%. By contrast with most genetic studies, we found variation between populations larger than between accessions. The largest between-population variation (46%) was for morphological traits. In particular, seed mass, leaf thickness and leaf width differed strongly between populations. Variation in growth characteristics between populations was poorly related to mean annual rainfall, mean humidity or January temperature at the sites of origin. We expect that differences between populations to be larger and correlation with environmental parameters stronger in plants grown in stressful conditions. According to our study, seed mass is more important than relative growth rate in determining variation in early plant biomass in *H. spontaneum*.

Key words: *Hordeum spontaneum*, intraspecific variation, leaf morphology, photosynthesis, relative growth rate, respiration, seed mass, variance components.

INTRODUCTION

Barley (*Hordeum vulgare*) is an important crop species often grown in areas with low rainfall, where crops such as wheat fail (Whabi & Gregory, 1989). The progenitor of this crop species is wild barley, *Hordeum spontaneum*, a diploid, self-pollinating annual that harbours large genetic variation (Brown *et al.*, 1978a; Nevo *et al.*, 1979, 1986; Corke *et al.*, 1988; Dawson *et al.*, 1993; Gunaskera *et al.*, 1994; Petersen *et al.*, 1994). As the rich gene pool of *H. spontaneum* can be used to improve cultivated barley, variability in this species is of interest to plant breeders and geneticists. Because of its occurrence in a wide range of different habitats (Nevo *et al.*, 1979), it is also interesting from an ecophysiological perspective.

Most research has focused on genetic differences between populations of wild barley, using isozyme polymorphisms, RFLP-markers and RAPD-markers. Large variation has been found between and within populations of *H. spontaneum* from different sites in Israel. The variation in polymorphisms of allozymes and disease resistance can be related to their natural environment (Nevo *et al.*, 1979). Nevo *et al.* (1984) also studied variation in quantitative traits of agronomic importance, such as

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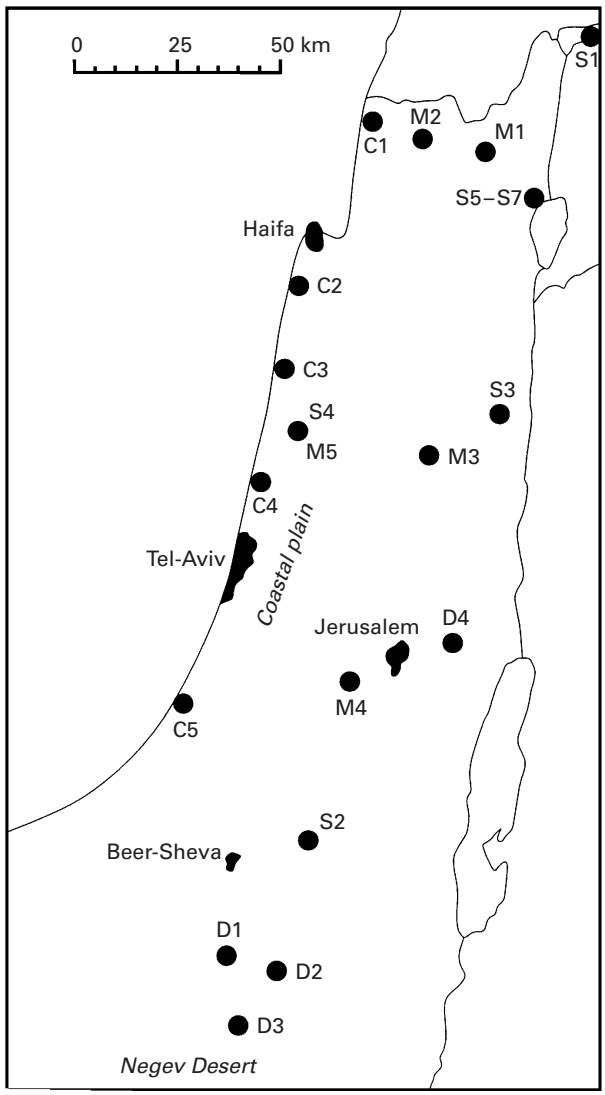

Fig. 1. Geographic distribution of sampling localities of *Hordeum spontaneum* in Israel. The characters refer to the different populations in Israel: M, mountain; C, coastal plain; S, steppic/marginal: D, desert.

vegetative biomass and number and mass of spikelets, spikes and stems. When grown under favourable conditions in a garden experiment, biomass of populations from mesic sites was about twice that of populations from xeric sites. It is therefore of interest to study the growth physiology of *H. spontaneum* and to investigate the causes of differences in biomass among plants from different habitats.

Differences in biomass can result from differences in seed mass, emergence time or variation in RGR (Van Andel & Biere, 1990). Differences in RGR have been found among species from different habitats; those in favourable environments have an inherently high RGR, whereas those from less favourable habitats have an inherently low RGR, even when grown in the same favourable conditions (Grime & Hunt, 1975; Poorter & Remkes, 1990). This does not necessarily imply that RGR has been the target of natural selection. It might well be that characteristics that are linked with or underlie RGR have been selected for (Lambers & Poorter, 1992; Poorter & Garnier, 1999). Differences in environment such as rainfall, resource availability, altitude or temperature are correlated with RGR and its components (Lambers & Poorter, 1992). In particular, Villar *et al*. (1998) found that *Aegilops* species from locations with a high annual rainfall have a higher RGR and invest less biomass in roots and more in shoots than species from drier locations.

Seed mass can also cause differences in biomass. In a study to determine the major factors responsible for variation in early vigour in barley, wheat and oat, embryo size was found the most important (López-Castañeda *et al*., 1996). Jurado & Westoby (1992) concluded that, among 28 native species from central Australia, seed size is more important than RGR or germination rate in determining seedling size 10 d after imbibition.

A comparison of strongly contrasting species has the advantage that differences in RGR and environment are generally large, but further genetic analysis is impossible. Thus, we chose to work with a single species occupying different habitats, concentrating on genetic differences in growth and growth components in favourable conditions as part of a larger study of the relationship between genetics and growth. We compared 21 different populations of *Hordeum spontaneum* from Israel, and four accessions (seed families) from each population. We asked the following questions:

- To what extent do the growth characteristics of these populations differ?
- How much of the variation can be explained by differences between populations, between accessions and within accessions?
- Do populations that are environmentally related show similarity in growth characteristics?
- Do differences in rainfall, humidity or temperature explain inherent variation between populations?
- Are differences in vegetative plant biomass under nonlimiting conditions of water and nutrient supply caused by differences in seed mass or differences in maximum relative growth rate?

MATERIAL AND METHODS

Plant material and growth

We studied 21 wild barley (*Hordeum spontaneum* C. Koch) populations from several locations in Israel (Fig. 1) which represent a wide range of geographical and environmental conditions. The populations are listed in Table 1 with climatic data. For ease of reference we divided the populations, somewhat arbitrarily, into four ecogeographical groups: three Mediterranean (mountain (M), coastal plain (C) and steppic (S)) and one from the desert (D). Three populations from Tabigha were sampled, two (Tab-

Table 1. *Location of the* Hordeum spontaneum *populations from Israel used for the analysis, and selected environmental data of sites of origin based on Nevo* et al. (*1984*); *Pakniyat* et al. (*1997*)

No.	Site of origin	Longitude (°E)	Latitude (°N)	Altitude (m)	Mean temperature (°C)			Mean annual rainfall (mm)	Mean humidity at 14:00 hours (%)
					Annual	Aug	Jan		
Mediterranean									
Mountain									
M_1	Mt. Meron	35.40	33.05	1150	14	22	6	1010	49
M_2	Maalot	35.27	33.00	500	17	23	8	790	50
M_3	Shechem	35.23	32.23	400	18	24	9	620	46
M_4	Bar-Giyyora	35.08	31.72	760	17	26	10	540	49
M_5	Nahal Oren (N)	35.02	32.43	75	19	24	11	690	59
Coastal Plain									
C_1	Akhziv	35.10	33.05	10	20	26	12	620	60
C_2	Atlit	34.95	32.70	50	20	26	13	500	65
C_3	Caesarea	34.90	32.50	10	20	26	13	540	65
C_4	Herzliyya	34.80	32.17	25	20	26	13	530	65
C_5	Ashqelon	34.60	31.63	50	20	27	14	420	64
Steppic/Marginal									
S_1	Mt. Hermon	35.75	33.28	1530	11	20	1	1600	52
S_2	Tel Shoket	34.92	31.32	375	19	26	11	280	45
S_3	Mehola	35.48	32.13	−150	22	30	13	270	34
S_4	Nahal Oren (S)	35.02	32.43	75	19	24	11	690	59
S_5	Tabigha	35.53	32.90	0	24	32	15	440	45
S_6	Tabigha (terra rossa)	35.53	32.90	0	24	32	15	440	45
S_7	Tabigha (basalt)	35.53	32.90	0	24	32	15	440	45
Desert									
D_1	Revivim	34.75	31.02	320	20	27	10	130	38
D_2	Yeroham	34.90	30.98	490	19	26	10	130	35
D_3	Sede Boqer	34.78	30.87	450	19	26	9	90	36
D_4	Wadi Qilt	35.38	31.83	50	23	30	14	170	40

TR and Tab-B) from a 100-m transect along an edaphic cline, one from terra rossa soil (derived from Middle Eocene hard limestone) one from basalt soil (generated on Pleistocene basalt flows, respectively (Nevo *et al.*, 1983). Two populations from Nahal Oren were sampled, one from the hot sunny south-facing slope, one from the cooler more humid north-facing slope (Nevo *et al.*, 1997).

Seeds were germinated on moistened filter paper in Petri dishes in a refrigerator at 6°C and an irradiance of 10 μmol m^{-2} s^{-1}. After 1 wk, seedlings were transferred to a container with drainage holes which was filled with clean white beach sand. The sand was saturated with half strength of the following nutrient solution: 603 μM $Ca(NO_3)_2$, 795 μM KNO_3, 190 μM KH_2PO_4, 270 μM $MgSO_4$, 0.2 μM $MnSO_4$, 0.9 μM $ZnSO_4$, 20 μM H_3BO_3, 0.3 μM Na_2MoO_4, 40 μM Fe-EDTA, 40 μM $FeSO_4$ and 47 μM SiO_2. The container was placed in a growth room for 5 d in the following conditions: 14:10 h day:night; 20°C day:night; irradiance of 450 ± 25 μmol m^{-2} s^{-1}; 70% rh. Thereafter seedlings were transferred to 33-l tanks containing the nutrient solution already described, aerated and at full strength, which was replaced weekly. The pH of the nutrient solution was adjusted regularly to 5.8 with H_2SO_4. To avoid mutual shading, the number of plants on each container ranged from 18 to 6, depending on size. Plants were rotated four times a week within the growth room.

Experimental design

Of each population, plants from four accessions (the progeny group from a single plant collected in the field) were grown. Of each accession eight plants were used to measure four sets of traits: allocation-related, physiological, chemical and morphological. Plants were measured 23–25 d after germination, when there were 2–10 tillers. Whole-shoot photosynthesis and shoot and root respiration were measured on two plants of each accession. Fresh and dry mass of leaves, stems and roots, leaf area, leaf width, leaf angle, shoot height and number of leaves and tillers were also determined on these two, and on two additional, plants. Two other plants were used for measurement of osmotic potential and to determine the chlorophyll concentration, and the remaining two plants were used for measurement of leaf thickness. The latter four plants were also used for chemical analyses. Because of the large number of plants and the time needed for measurement,

Table 2a. *Mean, P10 (10th percentile)-, P90 (90th percentile)- values, units of allocation-related and physiological traits as well as the percentage of variation per trait explained by differences between populations (df = 20), between accessions (df = 63) and within accessions (df = 84–252) for* Hordeum spontaneum *populations*

						Variance components		
Traits	Mean	P10	P90	Unit	Coefficient of variation (%)	Between populations (%)	Between accessions (%)	Within accessions (%)
Physiological								
Relative growth rate	300	250	350	mg g^{-1} d^{-1}	13	**12**	10	79
Photosynthesis per unit leaf area	15.4	13.3	17.7	μmol m^{-2} s^{-1}	12	**16**	12	72
Photosynthesis per unit leaf mass	550	510	600	nmol g^{-1} s^{-1}	9	**12**	8	80
Shoot respiration	47	39	53	nmol g^{-1} s^{-1}	12	6	15	78
% Respiration	40	33	48	%	17	5	**24**	71
Root respiration	65	44	86	nmol g^{-1} s^{-1}	28	**31**	0	69
Transpiration	1.64	1.20	2.09	mmol m^{-2} s^{-1}	26	**15**	0	85
Water use efficiency	7.5	5.5	9.6	mg g^{-1}	21	**23**	10	67
Allocation-related								
Water content of leaf	6.9	6.0	7.7	g g^{-1}	10	**20**	**11**	70
Water content of stem	9.4	8.4	10.5	g g^{-1}	9	**20**	**30**	50
Water content of root	11.0	9.5	11.9	g g^{-1}	44	1	1	98
Leaf area ratio	16.7	14.3	19.3	m^2 kg^{-1}	12	9	**36**	55
Specific leaf area	35.2	30.3	40.7	m^2 kg^{-1}	11	5	**33**	62
Leaf mass fraction	0.47	0.44	0.51	g g^{-1}	6	**35**	**22**	43
Stem mass fraction	0.20	0.17	0.22	g g^{-1}	10	**18**	**36**	45
Root mass fraction	0.33	0.29	0.37	g g^{-1}	11	**41**	**29**	29

Percentages printed in bold indicate that the between accession or between population variation was significant in ANOVA ($P < 0.05$).

germination and growth of plants were staggered. Four randomly chosen accessions of different populations were measured each week.

Measurements

Morphological traits. Seeds were not dried before the experiment and contained approx. 6% water. Mass of each seed (coated caryopsis, without awn or spikelet stalk) was separately determined before germination with a Sartorius R160P (Gottingen, Germany) balance. Leaf thickness and epidermal thickness were determined microscopically at five points at the middle of the youngest fully grown leaf: on the main vein, on the fourth vein to the left and right of the main vein and between the fourth and the fifth vein to the left and right of the main vein. Average leaf and epidermal thickness were then calculated. Leaf width was taken as the average of measurements taken at five points in the middle of the youngest fully grown leaf. Leaf angle was determined by measuring the angle between the horizontal plane and the middle part of each of the four oldest fully grown leaves; thus that of leaves with a vertical orientation was 90°.

Physiological traits. Net photosynthesis and dark respiration were measured as CO_2 exchange. Intact plants were placed in a cuvette, with shoots and roots in separate compartments (Poorter & Welschen, 1993). The root compartment was filled with a continuously aerated nutrient solution, similar to that supplied to the plants in the tanks. Irridiance was similar to that in the growth room. CO_2 and H_2O exchange were measured differentially with infrared gas analysers (ADC, model 225 MK3, Hoddesdon, UK) after 2 h equilibration. Calculations of all gas-exchange parameters were made according to Von Caemmerer & Farquhar (1981). In this way, whole-plant photosynthesis per unit leaf area (PS_a), per unit leaf mass (PS_m), shoot respiration (SR) and root respiration (RR) were assessed.

Allocation-related traits. Total leaf area was determined using a Li-3100 area meter (LI-COR, Lincoln, NE, USA). Leaf area, and fresh and dry mass of the leaves (leaf blades), stems (leaf sheaths) and roots were determined to calculate water content (fresh mass–dry mass/dry mass) of leaf, stem and root (WC_L, WC_S, WC_R, respectively), leaf area ratio (LAR, leaf area per total plant dry mass), specific leaf area (SLA, leaf area per leaf dry mass), leaf mass fraction (LMF, leaf dry mass per total plant dry mass), stem mass fraction (SMF, stem dry mass per total plant dry mass) and root mass fraction (RMF, root dry mass per total plant dry mass). Dry mass

Table 2b. *Mean, P10 (10th percentile)-, P90 (90th percentile)- values, units of chemical and morphological traits as well as the percentage of variation per trait explained between populations (df = 20), between accessions (df = 63) and within accessions (df = 84–252) for* Hordeum spontaneum *populations*

Traits	Mean	P10	P90	Unit	Coefficient of variation (%)	Variance components		
						Between populations (%)	Between accessions (%)	Within accessions (%)
Chemical								
Chlorophyll content	430	350	520	$\mu mol\ m^{-2}$	13	**45**	**17**	38
Osmotic potential	−1.46	−1.65	−1.27	MPa	10	**18**	**17**	65
Mineral concentration	180	158	202	$mg\ g^{-1}$	10	**27**	**44**	28
Organic acid conc.	41	26	53	$mg\ g^{-1}$	23	**13**	**21**	66
Nitrogen conc.	65	60	69	$mg\ g^{-1}$	5	**28**	**28**	44
Carbon conc.	402	390	414	$mg\ g^{-1}$	3	**22**	**21**	57
Nitrate conc.	73.5	59.1	89.8	$mg\ g^{-1}$	18	**27**	**45**	28
Organic nitrogen conc.	48.1	43.7	51.5	$mg\ g^{-1}$	7	**38**	**11**	51
C:N ratio	6.2	5.8	6.7	$g\ g^{-1}$	6	**27**	**40**	33
Morphological								
Seed mass	36.3	18.8	48.4	mg	30	**53**	**11**	36
Leaf mass density	89	73	107	$g\ mm^{-3}$	17	**30**	**41**	29
Leaf angle	30.7	0	62.8	°	76	**35**	**26**	39
Leaf thickness	340	270	400	µm	14	**61**	**10**	29
Leaf width	9.8	6.6	12.5	mm	23	**59**	**29**	11
Thickness of epidermis	66	54	76	µm	13	**36**	**15**	49

Percentages printed in bold indicate that the between accession or between population variation was significant in ANOVA ($P < 0.05$).

was measured after plant material had been dried for 48 h at 70°C.

Chemical traits. The osmotic potential was determined on leaf samples from the middle (sample size approx. 30-mm) of three leaves from one plant, which were stored in sealed plastic bags at −20°C. The osmotic potential of the leaf sap was measured using a Wescor (Logan, UT, USA) Vapour Pressure Osmometer, model 5100 C. The chlorophyll concentration of the leaf was determined according to Lichtenthaler & Wellburn (1983) after extraction with 80% acetone. To determine the concentration of C, total N, NO_3^-, organic N, organic acids and minerals, plants of each accession were combined into two independent samples. The C and N concentrations of the samples were quantified with two elemental analysers (Carlo Erba 1106 and Carlo Erba 1110, Milan, Italy). Ash and ash alkalinity were determined as described by Poorter & Villar (1997). Results were used to calculate organic acid and mineral concentrations. The nitrate concentration, quantified according to Cataldo *et al*. (1975) was subtracted from total N to determine the organic N concentration.

Calculations and statistical analyses

Relative growth rate (RGR) was estimated, on the basis of allocation-related and physiological traits, using the formula given in Poorter & Pothmann (1992):

$$\mathrm{RGR} = \begin{bmatrix} PS_a \times SLA \times LMF \\ -SR \times (LMF + SMF) \\ -RR \times RMF \end{bmatrix} / C \qquad \text{Eqn 1}$$

(PS_a, photosynthesis per unit leaf area; SLA, specific leaf area; LMF, leaf mass fraction; SMF, stem mass fraction; SR, shoot respiration; RR, root respiration; RMF, root mass fraction; C, C concentration of the plant biomass). We only determined the C content of the leaves, assuming that it is representative of that of the whole plant. In reality, the C content of roots and shoots tend to be slightly lower than in the leaves (Poorter & Bergkotte, 1992), but these differences are not likely to affect the RGR calculation. A second assumption is that the rates of photosynthesis and respiration can be integrated over 24 hr. All parameters from the RGR formula were measured on the same day.

Data were analysed with SPSS for Windows (release 8.0; SPSS Inc., Chicago, IL, USA). Analyses of Variance (ANOVAs) were used to determine whether there were differences in the measured traits between populations and between accessions within populations. Variance components were calculated from the mean sum of squares, derived from a nested ANOVA (Sokal & Rohlf, 1981). A discriminant analysis was carried out to separate the 21 populations. To compute the discriminant score all variables in the analysis were standardized. This means that, over all cases, the score from one function will have a mean of zero and a standard deviation of one. Relations between the various traits

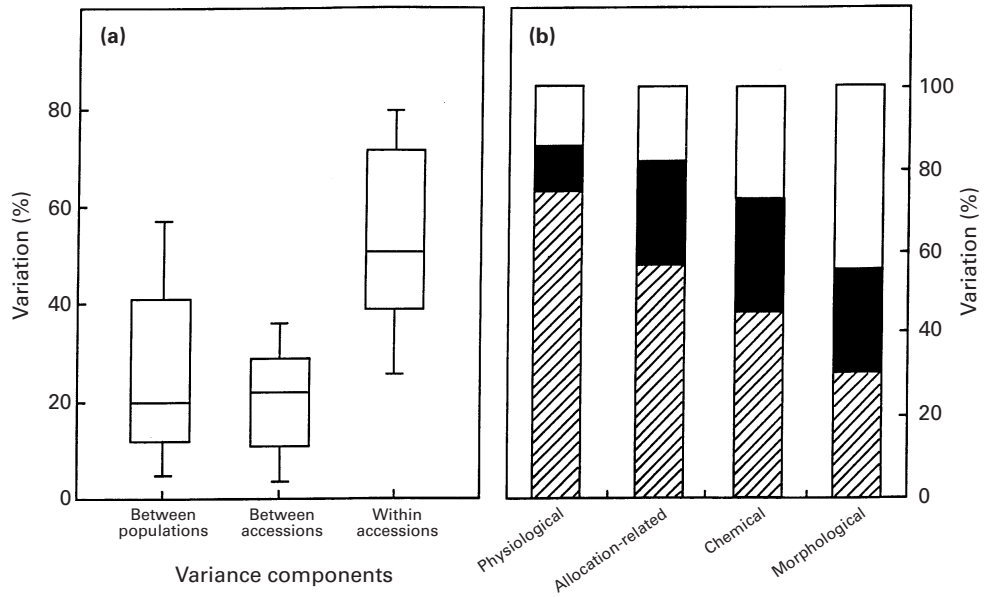

Fig. 2. (a) Average percentage of variation explained by the variance components, for all 31 traits. The box shows 50% (median), the range of the 25% and 75% quartiles. The error bars show the 10% and 90% borders. (b) Percentage of total variation explained by the variance components (average values), for the four groups of measured traits. Variations: open bar, between populations; closed bars, between accessions; hatched bars, within accessions.

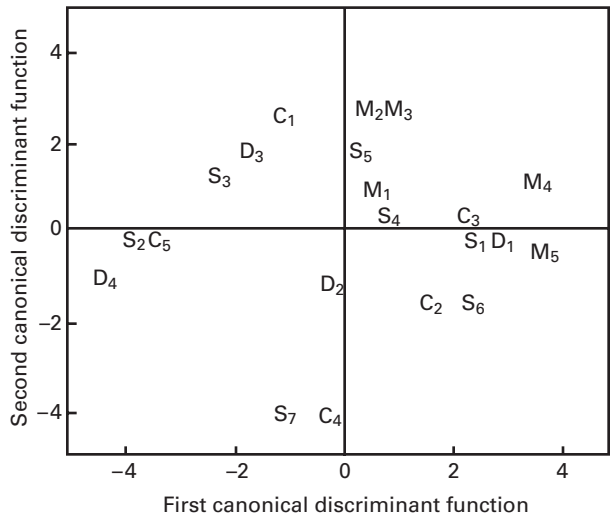

Fig. 3. Discriminant analysis of variables from Eqn 1 (see text for details; RGR, photosynthesis per unit leaf area, shoot respiration, root respiration, leaf mass fraction, stem mass fraction, root mass fraction, specific leaf area, C content) and morphological variables (seed mass, leaf thickness, leaf width) for populations of *Hordeum spontaneum*. The first discriminant function explains 37% and the second discriminant function explains 21% of the total variance. For explanation of characters see Table 1.

and the environmental data are described with linear multiple regression analyses.

RESULTS

Variation between populations, between accessions and within accessions

To give an indication of the observed values of all the measured traits in *H. spontaneum* an overview of the average over all populations and the P10 (10th percentile), the P90 (90th percentile) values as well as the coefficient of variation for each trait is given in Table 2. This table also lists the percentage variation explained by differences between populations, between accessions and within accessions for each trait separately as well as the significance of the variance components. The variables are subdivided into four groups of growth characteristics; physiological traits and allocation-related traits listed in Table 2a, and chemical traits and morphological traits listed in Table 2b. The division of the variables is somewhat arbitrary, but are made to facilitate comparison of sets of traits. The coefficient of variation shows the variability of all the traits. For 23 out of the 31 traits,

Table 3. *Largest absolute correlation between each variable and any discriminant function*

	Function	
Trait	1	2
Seed mass	0.66	−0.32
Leaf thickness	0.47	0.65
Root mass fraction	0.23	−0.10
Leaf width	0.18	0.52
Relative growth rate	0.05	−0.18
Photosynthesis per unit leaf area	0.03	−0.26
Shoot respiration	0.01	−0.02
Specific leaf area	−0.07	0.03
Leaf mass fraction	−0.16	−0.03
Stem mass fraction	−0.18	0.19
Carbon concentration	−0.23	−0.15
Root respiration	−0.37	−0.08

Table 4. *Summary of multiple regression between traits* (*average per population*) *and environmental factors of 20* Hordeum spontaneum *populations*

	Rainfall	Humidity	Temperature (Jan)	R^2
Relative growth rate	0	0	0	0.22
Photosynthesis per unit leaf area	−	0	0	0.32
Shoot respiration	0	0	+	0.24
Water content of leaf	+	0	0	0.34
Water content of stem	+	0	0	0.34
Seed mass	0	+	0	0.40
Leaf width	++	0	0	0.57
Leaf angle	++	0	0	0.42

+, positive correlation; −, negative correlation; 0, no significant correlation; $P < 0.05$, ++; −−, $P < 0.01$.

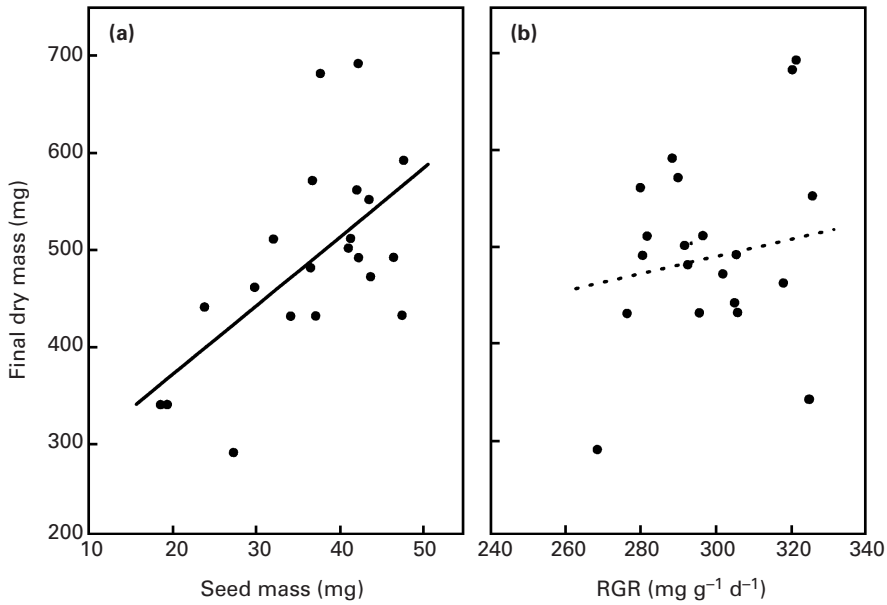

Fig. 4. Relation between the final plant biomass of *Hordeum spontaneum* and (a) relative growth rate ($R^2 = 0.02$) and (b) seed mass ($R^2 = 0.34$).

the coefficient of variation is <20%. Circa 80% of the variation in RGR and photosynthesis per unit leaf mass is due to differences within accessions, whereas only 12% of the variation is explained by differences between populations. By contrast, 59% and 61% of the variation in morphological parameters such as leaf width and leaf thickness respectively, is explained by differences between populations. Variation between populations was significant in all chemical and morphological variables and almost all physiological and allocation-related variables. The between-accession variation was significant for almost all allocation-related traits and all chemical and morphological variables but not significant for most of the physiological traits. Focusing on the between-population and between-accession variation, most variables, particularly the physiological and morphological variables, were found to have more variation between populations than between accessions (Tables 2a,b).

In summary, most of the variation in the measured traits was associated with differences within accessions rather than with differences between populations (Fig. 2a). This is particularly true for the physiological traits, where only 14% of the variation (Fig. 2b) is explained by differences between populations. The highest proportion of variance explained by differences between populations was observed for morphological traits (Fig. 2b).

Discriminant analysis

The discriminant analysis in Fig. 3 separates the populations in Israel based on the variables related to a plant's C economy (RGR, PS_a, SR, RR, SLA, LMF, SMF, RMF, C concentration) and three morphological variables, which differed the most between populations (seed mass, leaf thickness and leaf width). The independent variables were entered simultaneously. The first two functions explained 58% of the variation (Table 3). On the first function,

seed mass was the most important variable discriminating between populations. On the second function, leaf thickness was the most important trait. Morphological traits were more variable on the two functions than variables related to a plant's C economy, and RGR was neutral (Table 3). Of the C economy traits, biomass allocation to roots and leaves were the most important. Populations with a small seed mass are situated on the left side of the graph (Fig. 3). These include most of the desert populations. Almost all of the mountain populations are on the right side of the graph. The coastal and steppic/marginal populations are scattered within the graph.

Multiple regression between traits and environmental factors

Population S_1 (Mt. Hermon) was excluded from the multiple regression analysis between traits and environment, because this location not only has a very high rainfall but also a very high evaporation rate. Most of the investigated traits were not significantly correlated with an environmental factor at the site of origin. RGR was not related to any environmental factor (Table 4). PS_a was negatively correlated with rainfall and a positive relationship between SR and temperature was found. Water content of leaf and stem, as well as leaf width and leaf angle were positively correlated with rainfall. Seed mass had a positive relationship with humidity.

Seed size and final biomass

Seed mass varied by $>200\%$ between populations and total dry mass at harvest also by $>200\%$, and the two were positively correlated (Fig. 4a). There was, however, no significant correlation between the estimated RGR of the various populations and their final biomass (Fig. 4b). Therefore, most of the variation in total biomass of these 3.5-wk-old plants seems to be explained by variation in seed mass.

DISCUSSION

Variation between populations, between accessions and within accessions

In this study we analysed the variation between and within populations of *H. spontaneum*, grown under favourable conditions. Calculated over all plants investigated, almost 75% of all traits had a coefficient of variation $<20\%$. We expected higher variation, taking into account that *H. spontaneum* is an inbreeder (Brown, 1978a). Interestingly, other intraspecific studies on morphological variation in inbreeding species (Wolff, 1988; Bonin *et al.*, 1997) report similar coefficients of variation. For further analysis the variation in wild barley was divided into three variance components: between populations, between accessions and within accessions. Averaged over all parameters, variation within accessions explained 53% of the total variation. This is much higher than the values found for the variation between accessions within a population and between populations (21% and 26%, respectively). Maternal effects, environmental differences within the growth chamber or the influence of rare outcrossing rates (1.6% averaged over 26 populations; Brown *et al.*, 1978b), as well as stochastic factors could explain the large within-accession variation. The largest variation within accessions was observed for physiological traits and the smallest for morphological traits. A probable reason for this could be that the measurement errors involved in determining physiological variables are greater than for morphological variables.

In this study we calculated three variance components. However, when comparing these results with those from genetic variation studies in *H. spontaneum*, we have to consider that those studies often do not include the variation within accessions. This is because DNA is extracted from one plant only, or DNA samples are pooled, under the assumption that individuals within an accession are identical. This implies that the variation between accessions in those studies not only harbour the genetic variation, but also the variation within accessions. Table 2 shows that for most of the significant variance components, variation between populations is larger than within populations. This is contrary to many genetic diversity studies of allozymes and RFLPs in *H. spontaneum*, where most variation is explained by differences between accessions (Nevo *et al.*, 1986; Dawson *et al.*, 1993; Zhang *et al.*, 1993; Baum *et al.*, 1997). Most of these studies examined a greater number of accessions per population than our study, which might have resulted in a larger between-accession variation.

Photosynthesis expressed both per unit leaf area and per unit leaf mass (Table 2a), shows more variation between populations than between accessions, with the between-accession variation not significant. In contrast to these results, Carver & Nevo (1990) found in wild wheat (*Triticum dicoccoides*) populations that accessions within a given population showed 10-times more variation in PS_a or per unit chlorophyll than populations from different locations in a region. They concluded that genetic resources are located in relatively confined geographic regions.

Compared with the physiological and allocation-related variables, variation at the population level in chemical and morphological traits is larger (Table 2b, Fig. 2b). For all traits in the last two groups the variation between populations as well as the variation between accessions is significant. The largest between-population variation in the morphological category was the variation in leaf thickness (61%)

and in seed mass (53%). Corke *et al*. (1988) also found wide differences in chemical traits (N concentration in the leaf and stem) and in allocation-related traits (leaf and stem mass) between seven populations of *H. spontaneum*. In the present study most of the variation (48%) in chemical traits is explained by differences within accessions. In conclusion, *H. spontaneum* populations show greater variation in morphological variables than in physiological variables.

Discriminant analysis

The discriminant analysis (Fig. 3) based on all the variables of Eqn 1 as well as seed mass, leaf thickness and leaf width, showed that the morphological traits were more important than the physiological or allocation-related traits in discriminating populations (Table 3). Seed mass was the most important trait loading on the first canonical discriminant function (Table 3), separating desert from mountain populations. Desert populations had on average lower seed mass. One desert population (D_1) is at the right side of the graph, being morphologically different from the other desert populations. It is possible that barley plants at this site (Revivim), in the north-western Negev desert which is characterized by a high amount of dew, are not exposed to extremely dry conditions. Van Groenendael (1985) reports in a within species (*Plantago lanceolata*) study that plants from dry areas produce a large number of small seeds whereas plants from wet habitats produce fewer but bigger seeds. In contrast to these results are studies that make a comparison between species. Baker (1972) and Jurado & Westoby (1992) reported that species exposed to drought tend to have larger seeds and might therefore have larger root systems during early growth. On the second discriminant function (Table 3) leaf thickness was the most important trait. This function does not separate the populations very clearly, although it seems that most of the mountain populations have thicker leaves.

Multiple regression between traits and environmental factors

Most of the measured traits were not significantly correlated with an environmental factor at the site of origin. This might imply that the inherent differences between populations are not caused by adaptation to these environmental factors. However, one point of consideration is that the environmental data have not been measured directly at the sites of origin, but are averages from the nearest weather station. Another consideration is that we have grown plants under favourable conditions with an ample supply of nutrients and water. Under stressful conditions, differences in physiology and the relation to environmental factors will probably be greater. For example, Forster *et al*. (1997) found differences in experimentally determined abiotic stress responses (salt and drought tolerance) that are related to the environmental data of the sites of origin of *H. spontaneum* populations. The traits that showed a correlation with an environmental factor are listed in Table 4. Plants from xeric environments have a higher rate of PS_a than plants from mesic areas. The same result was found by Nevo *et al*. (1991) in wild wheat (*T. dicoccoides*). In the present study, plants from a xeric climate have relatively narrow leaves, a more horizontal position of the leaves (a lower leaf angle) and more tillers. Plants from xeric climates generally have smaller leaves, which might help in reducing transpiration (Von Willert *et al*., 1992). By contrast, leaves in a more horizontal position have a higher light absorption and therefore a higher transpiration rate. The horizontal position might also explain the higher rate of PS_a in xeric populations. Surprisingly, SLA, leaf thickness or LMF, usually determinants of photosynthesis, did not correlate with any of the environmental factors. This is at variance with the suggestions of, for example, Poorter & Garnier (1999) and Ehleringer (1981) that plants in drier environments have lower SLA. For *Aegilops* species, Villar *et al*. (1998) did not find a relationship between SLA and rainfall, but they did find a relation between LMF and rainfall.

Seed size and final biomass

Nevo *et al*. (1984) found a higher biomass in mesic *H. spontaneum* populations than in xeric populations, grown in a mesic habitat. We obtained similar results for 3.5-wk-old plants grown under near-optimal conditions. However, these differences in biomass are not caused by differences in RGR (Fig. 4b). Rather, differences in final mass could be explained by variation in seed mass (Fig. 4a). This is in agreement with Chapin *et al*. (1989), where seed size was found to be more important than RGR, 35 d after sowing, in determining plant size in different *Hordeum* species under favourable nutrition. The xeric populations produce smaller seeds, but there was no correlation between seed mass and RGR. Seed size usually correlates negatively with RGR in interspecific studies (Shipley & Peters, 1990; Jurado & Westoby, 1992; Marañón & Grubb, 1993; Reich *et al*., 1998). Meerts & Garnier (1996) studied seed size within a species (*Polygonum aviculare*) and found a positive relation between RGR and seed mass. Clevering (1999) studied also differences within a species (*Pragmites australis*), but found, like our study, no relation between RGR and seed size or between RGR and dry mass but a positive relation between seed size and dry mass. Similar suggestions were made by Van Andel & Biere (1990) and Garnier & Freijsen (1994). In the present study, plants

remained vegetative throughout the experiment. In the field between-populations differences in reproductive effort or length of the growing season (no data available) can be additional factors that determine differences in biomass.

CONCLUSIONS

Under the present favourable growth conditions, *H. spontaneum* populations from Israel show a larger between-population variation than in most genetic studies. Most of the between-population variation occurred in morphological traits. The differences are not strongly related to the natural habitat, though xeric and mesic sites can be distinguished based on morphological traits. Seed mass is an important trait for which *H. spontaneum* has large variation and was more important than RGR in determining vegetative biomass in *Hordeum* in our study.

ACKNOWLEDGEMENTS

The authors thank Peter van Tienderen and Maria Schipper for statistical support. We thank Avigdor Beiles, Arjen Biere, Ivonne Elberse, Eric Garnier, Bill Shipley, Piet Stam, Margreet ter Steege, Tytti Vanhala and Rens Voesenek for giving valuable comments on previous versions of this manuscript. Eviatar Nevo is grateful for financial support from the Israeli Discount Bank Chair of Evolutionary Biology and the Ancell-Teicher Research Foundation for Genetics and Molecular Evolution. The Earth and Life Science Foundation (ALW), which is subsidized by the Netherlands Organisation for Scientific Research (NWO), financially supported this study.

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