Developmental stability and adaptive radiation in the *Spalax ehrenbergi* superspecies in the Near-East

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Abstract

Chromosomal species of the mole rat, *Spalax ehrenbergi*, in Israel have been shown to display distinct adaptive strategies to increasing aridity. This adaptive radiation appeared to be associated with an increase in allozymic heterozygosity. In the present study, the developmental stability (DS) estimated by fluctuating asymmetry (FA) of dental traits was used to assess the suitability of habitat and the efficiency of adaptation to local environmental conditions among populations and chromosomal species. Although FA levels were highly heterogeneous among populations, they were not found to differ between species. DS of populations appeared, however, to be impaired at higher altitudes and in indurate soils. Since these environmental features were largely covariant, the effect of each one could not be precisely determined. Interestingly, while aridity is considered as the major selective force acting on populations southwards, DS was not altered under arid conditions, suggesting that mole rat populations were adapted to their local conditions of aridity. However, the cline of aridity is matched to several environmental and genetic clines among which are the increasing heterozygosity and recombination rate among species southwards. In studies of natural populations, the potential complementary effects of environmental and genetics on DS have to be considered and hamper the interpretation of habitat suitability expressed by DS in terms of adaptive strategies.

Introduction

The subterranean mole rat *Spalax ehrenbergi* superspecies in the Near-East is probably the most documented model of chromosomal speciation associated with an adaptive radiation (see Nevo, 1991, for review; Nevo *et al*., 1994a,b, 1995). Five chromosomal species present a southward trend of increasing chromosomal number in Israel (2n = 52, 54, 58 and 60I) and Northern Egypt (2n = 60E) (Fig. 1). Each of these species occurs in distinct climatic regions, the two extremes being represented by the cool and semihumid Golan heights (2n = 54) and the arid Negev desert (2n = 60I). Studies on numerous characters including physiological, ecological and behavioural traits have shown that each of the chromosomal species of Israel displays distinct adaptive strategies underlying the adaptive radiation into four climatic regimes associated with the chromosomal speciation process within this superspecies. Among the chromosomal species, the genetic diversity increases southwards, toward xeric environments (Nevo & Cleve, 1978; Nevo *et al*., 1996). More precisely, heterozygosity positively correlates with aridity stress, climatic unpredictability and increased steppic conditions (Nevo *et al*., 1994b, 1995). Recently, the chiasma frequency was shown to increase with chromosomal number among species (Nevo *et al*., 1996). It has been hypothesized that the adaptation of species to a more xeric environment at

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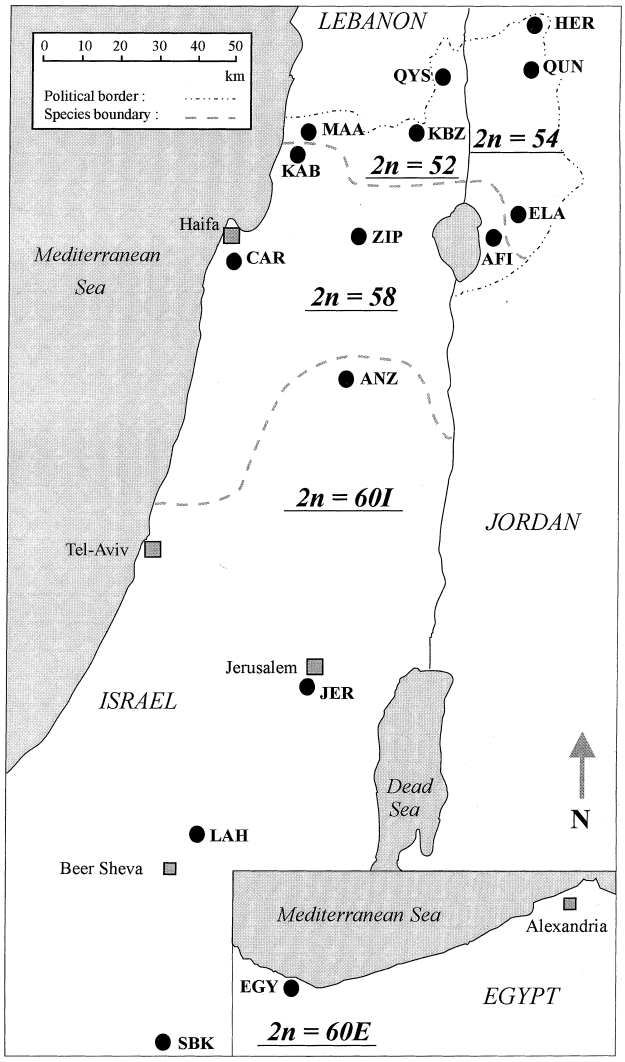

Fig. 1 Distribution of chromosomal species of *Spalax ehrenbergi* in Israel and Egypt and sample locations.

each step of increasing diploid number could be favoured by the higher level of genetic diversity amplified and maintained by the increase in recombination rates related to higher 2n. This phenomenon does not seem to be limited to this superspecies: *S. leucodon* studied in Turkey clearly exhibits the same trend of increasing diploid number associated with an increase in genetic diversity toward arid, contrasted and thus stressful environments (Nevo *et al.*, 1994b, 1995).

Responses to a new environmental stress at individual or population levels are required to avoid extinction or to allow the colonization of new areas characterized by different environmental features than those of the original range. Stress responses are complex and could result in several kinds of adaptation according to the types of selection induced (see Hoffmann & Parsons, 1991, for review). Apart from stress evasion, adaptation results from a selection for stress resistance (Hoffmann & Parsons, 1991) extending the tolerance of the species to more extreme habitats.

A relevant measure of habitat suitability in wild populations can be provided by the estimation of the level of developmental stability of morphological characters (Parsons, 1990a, b; Hoffmann & Parsons, 1991; Graham *et al.*, 1993). Developmental stability is a component of the developmental homeostasis which is defined as the ability of an organism to withstand genetic and environmental disturbances encountered during development, so as to produce a predetermined optimum phenotype (Zakharov, 1989). The other component of the developmental homeostasis is canalization, referring to the processes by which consistent phenotypes are produced despite the variability of genetic and environmental conditions. The developmental stability of organisms, often estimated by fluctuating asymmetry (FA) levels, i.e. the variability of the distributions of right-minus-left measurements of normally symmetrical bilateral characters, is known to be dependent on both genetic and environmental conditions (see Parsons, 1990a,b; Markow, 1995, for review). The impairing effect of environmental stresses, e.g. thermic and audiogenic stresses, protein deprivation or pollution, on developmental stability has been established on a wide variety of organisms (see Parsons, 1990a, for review). Concerning the genetic basis, it has been shown that the level of developmental stability of organisms is related to both genomic heterozygosity and coadaptation (Palmer & Strobeck, 1986; Clarke, 1993). The role of genomic coadaptation has clearly been established by studies of hybrid zones which reported that coadaptive gene complexes in hybrid populations were disrupted inasmuch as parental taxa were highly divergent (see Graham, 1992, for review). In contrast, the role of heterozygosity is still debated (see Clarke, 1993, for review). Several examples exist in wild populations (Kat, 1982; Vrijenhoek & Lerman, 1982) in which a cline of heterozygosity matches that of developmental stability. In these cases, the developmental instability is believed to be imputable to the high level of homozygosity subsequent to founder effects in marginal populations. However, Clarke (1993) suggested that it could rather result from the fixation of deleterious recessive alleles or from environmental stresses in these nonoptimal habitats and emphasized that, for developmental stability studies, the 'use of natural populations is fraught with danger unless there is a good understanding of the genetic structure and evolutionary history of the populations under examination.'

The evolutionary history of the *S. ehrenbergi* superspecies is well known and may be a relevant model to discuss the effect of genomic and environmental stresses on the developmental stability. In contrast with the examples cited above, in which heterozygosity levels in natural populations decrease towards marginal and

stressful environments, heterozygosity in the mole rat is believed to be a major component of adaptation to a more xeric and unpredictable environment southwards (Nevo & Beiles, 1988). In order to appraise the suitability of the various habitats of *S. ehrenbergi*, we estimated the level of developmental stability among several natural populations over the Near-Eastern range of the species. Additionally, the relationships between the level of developmental stability and several environmental features were studied to determine which of the latter may be considered as stressful. The quality and the efficiency of the distinct adaptive strategies displayed by populations and chromosomal species in response to their local environmental conditions – with special emphasis on the increase in aridity southwards – is then discussed with respect to the heterozygosity levels previously estimated for all localities considered in this study (Nevo *et al*., 1994a) as well as in the light of the evolutionary history of the chromosomal radiation.

Materials and methods

Samples

A total of 15 samples (Table 1) grouping 340 individuals belonging to the five chromosomal species of the *Spalax ehrenbergi* superspecies in Israel and Egypt were examined in this study. These animals represent all those available in the *S. ehrenbergi* collection of E. Nevo at the Institute of Evolution (University of Haifa). Several authors have underlined the biases related to the use of museum collection material for studies on developmental stability (Swaddle *et al*., 1994, 1995; Simmons *et al*., 1995). Particularly, pooling samples of different years or seasons may confound different environmental conditions. Swaddle *et al*. (1994) recommended comparing the subsamples of different years from a single locality before pooling them into a total sample. As for most subterranean rodents, *S. ehrenbergi* is difficult to capture and collecting an adequate sample size over the range of the superspecies in Israel and Northern Egypt requires several years of trapping. The samples considered in this study were trapped over 3–10 years (median: 6) from 1979 to 1991 and yielded too small sample sizes to test the year effect within localities.

Measurements

Fluctuating asymmetry was independently estimated on eight tooth measurements: the maximum length and width of the three lower molars (LM1, WM1, LM2, WM2, LM3, WM3) as routinely used in studies of FA in rodents (Bader, 1965; Alibert *et al*., 1994, 1997; Auffray *et al*., 1996) and the transversal (TI) and anterioposterior (API) diameters of the incisor. Molar measurements were taken using a Nikon measuroscope (0.001 mm) by S.R. and incisor measurements using a Mituyo calliper (0.01 mm) by J.-C.A. Some of the teeth were missing or broken depending on the preservation of skulls, which resulted in unequal sample sizes (Table 2). In *Spalax*, incisors are hypsodont, i.e. palliating wear, they grow during the lifetime of an individual. We do not have any evidence on the conservation of FA along the incisor. TI and API measurements were taken as close as possible to the incisor gap.

Measurement error appraisal was based on Palmer (1994). Using individuals from the locality ANZ, all measurements were taken twice during two different sessions. A mixed model ANOVA was performed for each trait considering the individual as the random effect, the side as the fixed one and their interaction. The significance of the interaction variance showed that the difference between sides varied more among individuals than would have been expected given the size of the measurement error (Palmer, 1994). Moreover, the results of these ANOVAS allow us to express the nondirectional asymmetry and the error of measurement in terms of the percentage of the total variance for each character. Error measurement was performed several months after the estimation of FA levels reported here. Although some additional individuals have increased the sample size for this locality, most incisors have been used for enamel analyses rendering them unavailable for appraisal of error. Consequently, the error estimation was based on 37 individuals for the first two molars, 23 for the third molar and 12 for incisor.

Statistical treatments

The FA is assessed by any estimator of the range of variability of the right-minus-left (R − L) distribution of a symmetrical bilateral character. However, this variability may not only represent the fluctuating asymmetry. Several preliminary tests have to be conducted before one may estimate and test levels of FA among characters and samples. The procedure we followed is largely based on Palmer & Strobeck (1986) and Palmer (1994) who exhaustively depicted the successive steps in the estimation and statistical comparison between FA levels among samples. Here, we have considered eight parameters and 15 samples leading to 120 distributions of (R − L) and consequently to 120 FA indices.

Normality assessment. Our samples cumulated several years of capture. Ross & Robertson (1990) have reported that some hybrid ant populations exhibit leptokurtic (R − L) distributions which may have resulted from pooling populations characterized by different levels of developmental stability (Graham, 1992). Without applying a sequential Bonferroni test, the normality of 23 out of the 120 distributions was rejected using the Dallal and Wilkinson approximation of the Lilliefors test (Sokal & Rohlf, 1995). Additionally, among the remaining normal distributions, 13 were shown to be skewed and/or leptokurtic. We considered this result as unsatisfactory

Table 1 Genetic and environmental variables for the samples of *Spalax erhenbergi*.

			Sample size		Genetic variables					Environmental features								
Locality	Code	2n	Fluctuating asymmetry	Genetic approach	A	P(1%)	P(5%)	H	HE	ALT	TM	TJ	TA	TD	RD	RN	HU	Soil
Israel																		
Qiryat shemona	QYS	52	10	14	1.306	0.278	0.111	0.035	0.047	187	19.0	9.5	26.0	16.5	60	655	49	Hard (Terra rossa)
Kerem-Ben-Zimra	KBZ	52	36	18	1.444	0.417	0.306	0.056	0.064	700	16.5	7.0	23.3	16.3	59	650	49	Hard (Basalt)
Maalot	MAA	52	23	13	1.389	0.361	0.139	0.039	0.049	500	16.8	8.1	23.2	15.1	53	785	53	Hard (Terra rossa)
Mount Hermon	HER	54	18	12	1.306	0.306	0.220	0.044	0.063	1300	12.4	3.0	20.1	17.1	65	1450	60	Hard (Terra rossa)
Quneitra	QUN	54	16	10	1.278	0.250	0.194	0.048	0.050	950	14.9	6.1	22.7	16.6	65	857	49	Hard (Basalt)
El Al	ELA	54	27	12	1.361	0.333	0.139	0.040	0.048	300	18.7	9.6	26.0	16.4	52	464	45	Hard (Basalt)
Kabri	KAB	58	20	14	1.500	0.417	0.361	0.058	0.088	100	20.0	10.5	26.0	15.5	50	600	58	Light (Rendzina)
Zippori	ZIP	58	16	15	1.556	0.412	0.333	0.076	0.094	250	18.5	10.4	26.0	15.6	53	500	48	Light (Rendzina)
Mount Carmel	CAR	58	17	15	1.472	0.472	0.389	0.098	0.121	400	17.0	11.5	24.0	12.5	55	720	58	Light (Marl)
Afiq	AFI	58	18	11	1.306	0.306	0.222	0.062	0.066	325	18.8	10.5	26.7	16.2	51	460	44	Hard (Basalt)
Anza	ANZ	60I	26	18	1.444	0.389	0.194	0.065	0.064	400	18.0	9.6	24.5	14.9	46	630	47	Light (Rendzina)
Jerusalem	JER	60I	28	18	1.667	0.556	0.417	0.087	0.115	700	17.5	8.7	23.9	15.2	42	500	51	Hard (Terra rossa)
Sede-Boker	SBK	60I	19	11	1.417	0.417	0.389	0.092	0.110	450	19.1	9.6	25.3	15.7	15	91	36	Light (Loess)
Lahav	LAH	60I	27	16	1.528	0.472	0.361	0.070	0.100	400	18.8	11.0	26.0	15.0	33	303	45	Light (Loess)
Egypt																		
EL Hamman	EGY	60E	38	17	1.250	0.250	0.194	0.029	0.038	32	20.2	14.4	26.9	12.5	29	184	63	Light (Loess)

A; Mean number of alleles per locus. P(1%); Mean proportion of loci polymorphic per population. The locus is considered as polymorphic if the proportion of the common allele is not greater than 0.99. P(5%); Mean proportion of loci polymorphic per population. The locus is considered as polymorphic if the proportion of the common allele is not greater than 0.95. H; Mean proportion of loci heterozygous per individual. HE; Gene diversity. Equal to expected heterozygosity under panmixis. ALT; Altitude in m. TM; Mean annual temperature in °C. TJ; Mean January temperature in °C. TA; Mean August temperature in °C. TD; Seasonal temperature difference °C. RD; Mean number of rainy days. RN; Mean annual rainfall in mm. HU; Mean humidity at 14:00 in %.

from a biological point of view but also from a statistical one, since we intended to test the population and species effects by applying ANOVAS. However, from a biological point of view, the rejection of normality for the (R − L) distributions might be more likely to be the result of the very heterogeneous levels of tooth wear observed on this material than of the pooling of samples exhibiting different directional asymmetry (DA) indices or antisymmetry (AS) patterns. DA occurs when one side of a bilateral character is systematically larger than the other, while in AS, which also corresponds to a systematic deviation from symmetry, the side that is larger varies at random among individuals (Palmer, 1994). Consequently, the test of Grubbs (1969; Sokal & Rohlf, 1995) for detecting outlier observations was applied to each of the 120 distributions. By using this procedure, 28 individual asymmetries were detected as outliers out of the total of 2377, i.e. only 1.2% of the whole data set, and these were excluded from further analyses.

Normality for each of the 120 resulting (R − L) distributions was tested using the Dallal and Wilkinson approximation of the Lilliefors test of normality (Sokal & Rohlf, 1995). Skewness and kurtosis were estimated and tested for all these distributions.

Directional asymmetry was tested within each of the 120 distributions of (R − L) by a *t*-test, the null hypothesis being the equality between the mean of distribution and 0.

Size dependence of FA was appraised within and among populations for each variable. The within-sample dependence was tested by the significance of the linear regression of |R − L| on (R + L)/2. Size dependence among samples was assessed by the linear regression between the mean of (R + L)/2 and log(var(R − L)).

During these preliminary treatments, numerous statistical tests were performed increasing the occurrence of type 1 error. A sequential Bonferroni technique would have been too conservative if, as proposed by Palmer (1994), it was applied over the 120 related statistical tests performed at each step of these preliminary treatments, i.e. appraisals of normality, skewness, kurtosis, directional asymmety, and size dependence within samples. Rather, and in order to establish independently the response of the traits at each step of this procedure, we conducted a Bonferroni test on each collection of $k = 15$ sample-related tests according to Rice (1989).

FA assessment and testing. Following the recommendations of Palmer (1994), two indices of FA, FA1 and FA4, were retained in this study. The FA1 index corresponds to the mean of the |R − L| distribution and is considered as being probably the most generally useful index for moderate to large sample sizes (Palmer, 1994). FA4 corresponds to the variance of the (R − L) distribution. This index is more sensitive to sample size but, in contrast to FA1, it is unbiased by DA.

The most appropriate way to test the differences of FA among chromosomal species and localities was to perform for each variable a two-level nested ANOVA on the |R − L| data sets used to calculate FA1. However, considering all traits together, we also performed a Levene's test as suggested by Palmer (1994). This test corresponds to a two-way model ANOVA performed on the total |R − L| data set, implying locality and trait as fixed effects and their interaction. To test the differences among species, the same procedure was applied but the locality effect was replaced by the chromosomal species one.

Finally, Kendall's coefficient of concordance was applied to test the concordance of FA indices, for FA1 and FA4, respectively, for the eight characters among populations. This allowed us to establish for each sample a synthetic ranking Rj of FA level over all traits which corresponds to the sum of the ranking each sample has obtained for all parameters.

Relationships between FA, genetic and environmental features were appraised among populations by correlation tests between these features and FA1 indices for each trait independently. Gamma correlation tests of rank (Siegel & Castellan, 1988) were also performed between each genetic or environmental variable and the sum of ranks Rj. The effect of the hardness of soils was tested by a Kruskall–Wallis analysis on each FA trait as well as on Rj, considering two groups of populations, those living in hard soils (basalt and terra rossa) versus those in light ones (rendzina, loess and marl). We also tested the relationships among samples between FA levels considered trait by trait and heterozygosity estimates considered locus per locus for 25 polymorphic loci as provided by Nevo *et al*. (1994a) for subsamples of those used in the present study (*Ada*, *Adk*, *αGpdh*, *Ald*, *Ap-1*, *Ap-2*, *Est-3*, *Est-4*, *Got-1*, *Got-2*, *G6pdh*, *Hk-2*, *Idh-1*, *Ldh-1*, *Mdh-1*, *Mdh-2*, *Me-1*, *Me-2*, *Mpi*, *Np*, *Pgi*, *Pgm-1*, *Pglm-2*, *Sdh*, *6Pgdh*).

When *P* values are not provided, they are encoded as follows: *: $P < 0.05$; **: $P < 0.01$; ***: $P < 0.001$.

Results

Preliminary treatments

Following the exclusion of outliers and the application of the sequential Bonferroni correction per traits, the normality was rejected for one distribution (trait TI for sample EGY). Also, three (R − L) distributions were skewed and leptokurtic (TI and LM2 for JER and TI for LAH) and one was leptokurtic (API for CAR) (Table 2). TI trait thus appeared often to depart from a normal distribution. However, we did not consider this as sufficient evidence to remove TI trait from the data set, but rather that it required special care.

None of the 120 means (R − L) statistically differed from 0 after having applied the sequential Bonferroni test (Table 2). Similarly, none of the 120 linear regressions of |R − L| on (R + L)/2 was significant, revealing the independence of size character and asymmetry within samples (Table 2). The *F* value of the linear regression of

Table 2 Detailed presentation of FA results follwing recommendations of Palmer (1994) (see Table 1 and text for Codes).

				(R + L)/2		Slope			(R – L)									IR – LI	
Species	Locality code	Traits	*n*	mean	SE	mean	SE	(*a*)	mean × 100	SE × 1000	(*b*)	Var (FA4) × 1000	Skew	SE	(*c*)	Kurtosis	SE	(*d*)	mean (FA1) × 100	SE × 100
2n = 52	QYS	API	10	2.45	0.053	0.056	0.095		–2.30	2.15		4.62	0.38	0.69		–0.96	1.33		5.30	1.45
		TI	10	1.84	0.032	–0.148	0.092		–3.40	2.02		4.09	0.14	0.69		–1.57	1.33		5.80	1.28
		LM1	8	2.48	0.049	–0.003	0.074		–1.38	1.69		2.30	–0.46	0.75		–0.86	1.48		4.05	0.89
		WM1	8	2.09	0.037	–0.014	0.090		–0.06	1.24		1.24	1.15	0.75		1.26	1.48		2.46	0.83
		LM2	7	2.20	0.045	–0.035	0.117		–2.76	1.83		2.35	–0.25	0.79		–0.78	1.59		4.39	1.19
		WM2	7	2.19	0.049	0.012	0.059		–0.67	1.06		0.79	–0.79	0.79		0.35	1.59		2.19	0.64
		LM3	4	2.09	0.046	0.097	0.027		–3.90	0.48	–	0.09	1.60	1.01		2.39	2.62		3.90	0.48
		WM3	5	1.88	0.084	0.018	0.087		–1.82	1.94		1.89	0.20	0.91		–2.56	2.00		3.46	1.27
		Rj										59							67	
	KBZ	API	30	2.48	0.041	0.016	0.031		–0.57	0.96		2.78	0.07	0.43		0.25	0.83		3.90	0.64
		TI	30	1.84	0.036	0.008	0.030		–0.73	0.79	*	1.87	0.85	0.43		0.27	0.83		3.07	0.56
		LM1	36	2.48	0.023	0.078	0.042		–0.78	0.84		2.55	0.19	0.39		0.96	0.77		3.65	0.59
		WM1	35	2.01	0.035	0.017	0.025		1.77	0.94		3.08	–0.01	0.40		–0.87	0.78		4.95	0.50
		LM2	36	2.18	0.020	0.028	0.053		0.66	0.95		3.27	0.36	0.39		0.81	0.77		4.42	0.60
		WM2	36	2.09	0.042	–0.026	0.018		–0.59	0.72		1.85	0.57	0.39		–0.26	0.77		3.32	0.46
		LM3	27	2.08	0.036	0.014	0.036		2.17	0.92		2.28	–0.19	0.45		1.30	0.87		3.98	0.65
		WM3	28	1.85	0.033	–0.051	0.037		–2.11	0.89		2.24	–0.02	0.44		1.24	0.86		3.86	0.64
		Rj										79							79	
	MAA	API	19	2.35	0.053	0.004	0.037		–1.79	1.21		2.76	–0.28	0.52		2.10	1.01		4.21	0.80
		TI	19	1.73	0.032	–0.015	0.035		–1.00	0.85		1.37	–0.14	0.52		–0.96	1.01		3.21	0.45
		LM1	20	2.43	0.023	0.032	0.041		–0.92	0.83		1.37	–0.71	0.51		–0.28	0.99		3.26	0.41
		WM1	20	2.01	0.027	–0.039	0.055		1.36	1.04		2.15	–0.12	0.51		–0.78	0.99		3.80	0.64
		LM2	22	2.10	0.018	0.033	0.107		–1.59	1.33		3.90	0.75	0.49		1.10	0.95		4.82	0.89
		WM2	22	2.09	0.026	0.062	0.039		0.31	0.83		1.51	–0.40	0.49		–0.17	0.95		3.11	0.48
		LM3	20	2.09	0.037	0.011	0.025		0.35	0.69		0.95	–0.56	0.51		–0.46	0.99		2.49	0.39
		WM3	20	1.84	0.026	–0.062	0.066		–1.07	1.20		2.87	0.75	0.51		–0.08	0.99		4.22	0.75
		Rj										62							66	
2n = 54	HER	API	17	2.54	0.043	0.136	0.064		–1.06	1.94		6.38	–0.32	0.55		0.49	1.06		6.12	1.22
		TI	17	1.90	0.031	–0.037	0.052		0.06	1.05		1.88	–0.89	0.55		0.40	1.06		3.12	0.71
		LM1	17	2.56	0.021	0.042	0.112		–3.30	1.54		4.03	–0.03	0.55		–0.67	1.06		5.99	0.90
		WM1	16	2.14	0.026	–0.019	0.076		–1.67	1.13		2.05	0.43	0.56		–0.62	1.09		3.74	0.73
		LM2	16	2.19	0.025	–0.043	0.055		–1.59	0.94		1.42	0.10	0.56		–1.46	1.09		3.43	0.52
		WM2	17	2.25	0.032	0.063	0.048		–0.99	1.17		2.31	–0.13	0.55		–0.39	1.06		4.04	0.64
		LM3	13	2.20	0.044	0.000	0.053		–2.39	1.04		1.40	0.28	0.62		–0.80	1.19		3.36	0.78
		WM3	13	1.98	0.035	–0.052	0.051		–0.42	1.01		1.33	–0.45	0.62		0.15	1.19		2.78	0.62
		Rj										68							70	
	QUN	API	16	2.68	0.054	0.058	0.049		–1.81	1.88		5.68	0.51	0.56		0.02	1.09		6.44	1.00
		TI	16	1.94	0.040	0.076	0.030		–0.38	0.93		1.37	–0.37	0.56		–0.49	1.09		2.88	0.56
		LM1	16	2.60	0.028	0.047	0.097		–4.31	1.58		4.01	–0.55	0.56		–0.26	1.09		6.36	1.02
		WM1	16	2.18	0.042	–0.014	0.077		1.49	1.82		5.32	1.01	0.56		1.75	1.09		5.44	1.22

		LM2	16	2.29	0.031	0.079	0.066	−0.21	1.25	2.50	−0.38	0.56	0.80	1.09		3.71	0.80
		WM2	16	2.26	0.049	−0.008	0.054	−3.17	1.26	2.55	0.54	0.56	−0.13	1.09		4.41	0.98
		LM3	16	2.28	0.033	0.005	0.059	0.59	1.23	2.44	0.40	0.56	0.13	1.09		3.90	0.73
		WM3	16	2.05	0.033	−0.030	0.035	−0.45	0.92	1.37	0.05	0.56	−0.93	1.09		3.15	0.45
		Rj								87						88	
-	ELA	API	24	2.61	0.048	0.037	0.033	1.00	1.26	3.81	−0.70	0.47	0.43	0.92		4.92	0.76
		TI	24	1.92	0.031	0.057	0.026	−0.21	0.70	1.19	−0.02	0.47	0.06	0.92		2.63	0.45
		LM1	25	2.60	0.026	−0.048	0.038	−1.34	0.80	1.61	−0.25	0.46	−0.28	0.90		3.42	0.48
		WM1	24	2.17	0.033	−0.031	0.055	1.68	1.22	3.55	−0.74	0.47	0.81	0.92		4.51	0.85
		LM2	23	2.27	0.019	−0.038	0.045	0.02	0.80	1.46	0.28	0.48	−0.70	0.93		3.21	0.41
		WM2	22	2.28	0.030	−0.071	0.032	−0.67	0.79	1.38	0.17	0.49	−0.27	0.95		2.97	0.48
		LM3	20	2.26	0.040	−0.009	0.030	−0.66	0.80	1.27	−0.80	0.51	0.83	0.99		2.78	0.50
		WM3	21	2.03	0.029	0.050	0.040	−2.00	0.90	1.69	−0.43	0.50	−0.66	0.97		3.83	0.52
		Rj								55						54	
2n = 58	KAB	API	19	2.34	0.063	0.036	0.033	0.32	1.21	2.80	0.61	0.52	2.14	1.01		3.58	0.88
		TI	19	1.74	0.053	0.035	0.023	1.79	0.78	1.16	0.27	0.52	0.53	1.01		3.05	0.52
		LM1	19	2.49	0.029	0.017	0.040	−1.09	0.68	0.88	0.49	0.52	0.06	1.01		2.34	0.48
		WM1	20	2.01	0.040	−0.013	0.032	0.58	0.85	1.46	−0.51	0.51	−0.48	0.99		2.94	0.54
		LM2	19	2.16	0.023	−0.032	0.041	−1.47	0.88	1.48	−0.27	0.52	−1.42	1.01		3.64	0.40
		WM2	20	2.09	0.042	0.013	0.024	0.94	0.65	0.86	0.23	0.51	−0.11	0.99		2.35	0.43
		LM3	20	2.15	0.044	−0.024	0.053	−1.96	1.38	3.80	0.50	0.51	0.27	0.99		4.65	0.98
		WM3	20	1.86	0.039	−0.042	0.051	−2.43	1.38	3.82	0.03	0.51	−1.09	0.99		5.26	0.87
		Rj								56						57	
	ZIP	API	14	2.32	0.060	0.024	0.031	1.07	1.12	1.75	1.29	0.60	1.38	1.15		3.50	0.63
		TI	15	1.69	0.037	0.043	0.038	0.33	0.90	1.21	−0.56	0.58	−0.50	1.12		2.73	0.53
		LM1	15	2.41	0.031	−0.006	0.064	2.79	1.06	1.67	0.20	0.58	−1.18	1.12		4.12	0.68
		WM1	15	1.98	0.028	−0.030	0.062	0.08	0.95	1.11	−0.73	0.60	1.25	1.15		2.68	0.63
		LM2	15	2.11	0.030	−0.009	0.081	−1.62	1.18	2.09	0.98	0.58	1.76	1.12		3.37	0.88
		WM2	15	2.07	0.025	0.043	0.039	0.97	0.73	0.80	0.20	0.58	−1.06	1.12		2.54	0.37
		LM3	13	2.14	0.039	−0.010	0.052	−0.85	1.21	1.91	−0.20	0.62	−0.61	1.19		3.60	0.67
		WM3	12	1.93	0.037	0.039	0.035	−0.10	0.90	0.98	−1.08	0.64	−0.04	1.23		2.62	0.44
		Rj								38						41	
	CAR	API	13	2.31	0.052	−0.031	0.070	1.15	1.60	3.33	−1.74	0.62	4.52	1.19	**	3.77	1.22
		TI	14	1.68	0.032	−0.017	0.062	0.14	0.89	1.11	−0.34	0.60	−0.10	1.15		2.29	0.62
		LM1	16	2.47	0.025	−0.015	0.064	−1.69	1.24	2.44	−0.36	0.56	−0.78	1.09		4.51	0.60
		WM1	14	1.97	0.036	−0.025	0.026	1.38	0.50	0.75	1.50	0.58	2.67	1.12		1.95	0.33
		LM2	16	2.09	0.019	−0.099	0.122	−0.31	1.30	2.69	0.75	0.56	1.86	1.09		3.66	0.89
		WM2	16	2.06	0.035	0.000	0.076	1.80	1.37	2.99	−0.65	0.56	0.39	1.09		4.05	0.99
		LM3	14	2.12	0.041	−0.102	0.062	1.43	1.25	2.20	−0.57	0.60	0.42	1.15		3.24	0.96
		WM3	13	1.88	0.041	0.033	0.036	−0.91	0.80	0.83	−1.50	0.62	2.90	1.19		2.31	0.51
		Rj								57						46	
	AFI	API	18	2.41	0.051	0.048	0.029	0.11	1.03	1.89	0.33	0.54	−0.71	1.04		3.33	0.63
		TI	17	1.76	0.044	−0.006	0.026	1.18	0.64	0.70	−0.92	0.55	0.13	1.06		2.00	0.50
		LM1	17	2.50	0.035	0.047	0.040	−1.95	0.94	1.51	−0.06	0.55	−0.37	1.06		3.60	0.56
		WM1	18	2.02	0.052	−0.035	0.045	2.04	1.44	3.73	−0.63	0.54	−0.60	1.04		4.89	0.95

Table 2 (continued)

Species	Locality code	Traits	*n*	(R + L)/2 mean	(R + L)/2 SE	Slope mean	Slope SE	(*a*)	(R − L) mean × 100	(R − L) SE × 1000 (*b*)	Var (FA4) × 1000	Skew	SE	(*c*)	Kurtosis	SE	(*d*)	IR − LI mean (FA1) × 100	IR − LI SE × 100
		LM2	18	2.20	0.027	0.107	0.040		−0.48	0.93	1.56	−0.10	0.54		−0.28	1.04		3.21	0.52
		WM2	18	2.12	0.053	0.041	0.036		−1.33	1.34	3.25	−0.56	0.54		0.47	1.04		4.66	0.80
		LM3	16	2.23	0.050	−0.045	0.063		−1.57	1.65	4.34	−0.04	0.56		0.93	1.09		4.67	1.19
		WM3	13	1.96	0.042	0.032	0.060		−2.79	1.44	2.71	−0.65	0.62		0.26	1.19		4.93	0.84
		Rj									71							68	
2n = 60 (Israel)	ANZ	API	23	2.49	0.042	0.034	0.029		1.52	0.84	1.63	−0.72	0.48		0.79	0.93		3.26	0.57
		TI	24	1.82	0.029	0.069	0.041		0.00	0.95	2.17	−0.01	0.47		0.04	0.92		3.58	0.59
		LM1	26	2.45	0.018	0.065	0.044		−0.32	0.70	1.29	−0.13	0.46		0.01	0.89		2.86	0.42
		WM1	26	1.93	0.033	0.032	0.043		1.24	1.04	2.82	−0.65	0.46		0.79	0.89		4.07	0.70
		LM2	26	2.16	0.018	0.004	0.044		−0.43	0.66	1.13	0.16	0.46		−0.35	0.89		2.73	0.38
		WM2	26	2.00	0.032	−0.019	0.031		0.58	0.73	1.39	1.05	0.46		1.85	0.89		2.80	0.48
		LM3	23	2.04	0.041	−0.063	0.039		0.19	1.26	3.67	1.50	0.48		2.41	0.93		4.39	0.85
		WM3	22	1.79	0.030	0.014	0.026		−1.28	0.58	0.73	−0.03	0.49		−1.56	0.95		2.49	0.34
		Rj									49							45	
	JER	API	23	2.36	0.029	0.117	0.047		1.09	1.21	3.35	−0.17	0.48		−0.30	0.93		4.74	0.70
		TI	23	1.78	0.048	0.055	0.041		0.57	1.16	3.11	−1.85	0.48	**	4.69	0.93	**	3.61	0.88
		LM1	23	2.41	0.025	−0.066	0.063		0.47	1.30	3.89	0.62	0.48		−0.22	0.93		5.03	0.74
		WM1	23	1.97	0.030	0.081	0.049		2.05	1.04	2.47	−0.40	0.48		0.88	0.93		4.13	0.70
		LM2	24	2.14	0.017	0.301	0.105		2.06	1.19	3.39	−1.82	0.47	**	5.48	0.92	**	3.75	0.99
		WM2	24	2.05	0.025	0.024	0.052		−0.66	1.11	2.93	0.69	0.47		0.28	0.92		4.43	0.62
		LM3	21	2.11	0.032	0.028	0.081		0.60	1.67	5.84	0.04	0.50		1.06	0.97		5.48	1.14
		WM3	21	1.82	0.024	0.061	0.091		−3.57	1.21	3.09	0.92	0.50		0.62	0.97		4.84	0.97
		Rj									101							100	
	SBK	API	15	2.40	0.081	−0.022	0.016		1.07	1.02	1.56	0.63	0.58		−0.26	1.12		3.47	0.52
		TI	15	1.83	0.056	−0.013	0.023		0.80	0.71	0.75	−0.41	0.58		−0.58	1.12		2.13	0.47
		LM1	18	2.44	0.034	0.042	0.069		−1.91	1.39	3.48	0.68	0.54		−0.23	1.04		4.63	0.94
		WM1	18	1.91	0.037	−0.010	0.050		−0.75	1.24	2.78	0.25	0.54		−0.70	1.04		4.17	0.74
		LM2	18	2.20	0.021	−0.083	0.088		−0.69	1.28	2.93	−0.08	0.54		−0.48	1.04		4.29	0.76
		WM2	18	2.05	0.034	0.019	0.054		0.66	1.26	2.84	0.50	0.54		−0.08	1.04		4.24	0.74
		LM3	13	2.06	0.034	−0.078	0.075		−0.68	1.31	2.25	1.21	0.62		1.03	1.19		3.40	0.90
		WM3	13	1.84	0.053	0.063	0.073		1.50	1.99	5.15	−0.67	0.62		0.99	1.19		5.38	1.32
		Rj									73							74	
	LAH	API	20	2.38	0.050	−0.005	0.029		−0.95	1.11	2.45	0.29	0.51		−0.29	0.99		3.95	0.67
		TI	20	1.80	0.040	−0.009	0.061		−1.05	1.18	2.79	1.96	0.51	**	4.01	0.99	**	3.35	0.93
		LM1	26	2.39	0.016	−0.005	0.076		0.12	0.99	2.57	0.30	0.46		−0.45	0.89		3.99	0.59
		WM1	24	1.91	0.029	0.050	0.027		1.50	0.66	1.04	0.19	0.47		−0.07	0.92		2.93	0.40
		LM2	24	2.10	0.017	−0.048	0.052		−1.37	0.66	1.05	−0.21	0.47		0.11	0.92		2.77	0.43
		WM2	24	2.00	0.029	0.005	0.040		−0.85	0.75	1.34	1.04	0.47		0.64	0.92		2.65	0.53
		LM3	14	2.08	0.049	−0.033	0.044		−1.31	1.11	1.74	−0.05	0.60		0.36	1.15		3.21	0.76
		WM3	15	1.85	0.031	0.131	0.049		−1.71	1.06	1.69	0.50	0.58		0.31	1.12		3.53	0.67
		Rj									49							50	

2n = 60 (Egypt)	EGY																	
		API	37	2.37	0.049	0.032	0.011		−0.05	0.57		1.21	0.74	0.39		0.61	0.76		2.70	0.35
		TI	37	1.75	0.036	0.032	0.011		−0.24	0.40	*	0.59	0.89	0.39		1.03	0.76		1.76	0.28
		LM1	34	2.56	0.022	0.124	0.049		0.04	1.09		4.04	0.11	0.40		−0.05	0.79		4.91	0.68
		WM1	34	1.88	0.019	0.010	0.034		0.39	0.63		1.33	0.12	0.40		−0.10	0.79		2.92	0.37
		LM2	35	2.20	0.011	0.084	0.081		0.47	0.81		2.32	−0.59	0.40		0.87	0.78		3.67	0.52
		WM2	34	2.06	0.022	0.057	0.042		1.63	0.82		2.27	−0.15	0.40		0.09	0.79		3.96	0.52
		LM3	28	2.19	0.017	−0.067	0.069		−0.37	1.08		3.26	−0.32	0.44		0.02	0.86		4.62	0.61
		WM3	28	1.87	0.021	0.008	0.041		−1.39	0.76		1.61	−0.27	0.44		−0.75	0.86		3.51	0.43
		Rj										56							55	

(a) Significance of F-Value (Regression of |R−L| on (R + L)/2). (b) Significance of the normality test (Dallal and Wilkinson approximation to Lilliefors test). (c) Significance of Skewness. (d) Significance of Kurtosis. – no tested (small sample size). $^*P < 0.05$; $^{**}P < 0.01$.

log(var(R − L)) on mean((R + L)/2) was significant only for API ($F = 5.58$, d.f. 1,13, $P = 0.034$). Applying the Fisher method for multiple independent tests (Manly, 1985) ($\chi^2 = 17.5$, d.f. 16, $P = 0.35$), no significant size dependence of FA among traits was shown.

The interaction term of mixed model ANOVAS to test the error of measurement was highly significant for all traits ($10^{-29} < P < 10^{-5}$) indicating that the difference between sides varied much more among individuals that would have been expected given the measurement error. Over all traits and on the subsample used for the estimation of measurement error, the nondirectional asymmetry explains between 1.61% (trait API) and 5.66% (LM1) of the total variance, while the error of measurement accounts for 0.14% (API) and 1.5% (LM1).

FA levels among populations and chromosomal species

Estimates of FA1 and FA4 for all localities are provided in Table 2. The unbalanced and two-level nested (locality within chromosomal species) ANOVA on the |R − L| data set indicated that FA levels were strongly heterogeneous among localities (Table 3). The chromosomal species effect, even if significant for the trait API, may not be considered as significant over all traits (after applying the Fisher method for combining independent tests), whereas the population effect remained highly significant (Table 3).

The Levene's test using locality and trait has shown that these effects and their interaction were significant (locality: $F = 2.95$***, d.f. 14,2257; trait: $F = 4.24$***, d.f. 7,2257; interaction: $F = 1.48$***, d.f. 98,2257). When the species effect replaced the locality effect, it was not significant (species: $F = 2.37$ ns, d.f. 4,2345; trait: $F = 4.62$***, d.f. 7,2345; interaction: $F = 2.32$ ns, d.f. 28,2345).

Additionally, Bonferroni–Dunn tests performed on each pair of localities showed that some of the populations exhibiting the highest levels of FA (QUN (2n = 54) and JER ($2n = 60$I)) were significantly different from those exhibiting the lowest levels (LAH (2n = 60I), ZIP (2n = 58), CAR ($2n = 58$), ANZ ($2n = 60$I) and EGY (2n = 60*E*)). The pattern of significantly differing populations showed no species-related pattern of developmental stability.

FA levels among traits

The significance of the trait effect at the preceeding step of the treatments indicated that some traits were better than others for revealing differences among localities. The plot of FA levels established over the all-individual dataset (not shown here) clearly indicated that FA exhibited by the TI trait was the lowest. It remains difficult, however, to relate the relatively important level of statistical

Table 3 Nested ANOVAS on $|R - L|$ data sets per trait (Fisher Method: see text; Fs′, d.f′. see Sokal & Rohlf, 1995).

	Species				Locality			
Traits	d.f. num	d.f.′	Fs′	*P*	d.f. num	d.f.	Fs	*P*
API	4	3.8	10.55	0.02	10	283	0.62	0.79
TI	4	6.7	2.27	0.17	10	285	1.29	0.23
LM1	4	8.2	0.99	0.46	10	301	2.34	0.01
WM1	4	7.7	1.23	0.37	10	296	1.82	0.06
LM2	4	2.7	5.71	0.11	10	300	0.46	0.91
WM1	4	8.0	0.27	0.89	10	300	2.01	0.03
LM3	4	7.5	0.61	0.67	10	299	1.59	0.11
WM3	4	8.4	0.09	0.98	10	300	2.59	0.01
Fisher method:			$\chi^2 = 20.12$ $P = 0.21$				$\chi^2 = 40.13$ $P = 0.0007$	

rejection for normality, skewness and kurtosis exhibited by this trait with its general lower level of FA.

Kendall's tests of concordance have revealed that the rankings of populations based on FA1 were not independent among traits (W = 0.22, $\chi^2 = 24.38$, d.f. 14, $P = 0.041$) but this pattern remained slightly nonsignificant using FA4 (W = 0.21, $\chi^2 = 23.01$, d.f. 14, $P = 0.060$). However, when the TI trait, which exhibited the lowest level of FA established over the all-individual data set, was excluded from this analysis, the dependence among the remaining traits strongly increased using both FA1 (W = 0.27, $\chi^2 = 30.35$, d.f. 14, $P = 0.007$) and FA4 (W = 0.26, $\chi^2 = 27.53$, d.f. 14, $P = 0.016$). The global significance of the Kendall test of concordance conducted on the eight original traits allowed us to consider the resulting sum of ranking per population as a synthetic rank (Rj) of FA level obtained by each population over the eight traits (Table 2). FA was assessed by two indices, and thus two synthetic rankings were obtained, respectively, based on FA1 and FA4. The rank correlation between these two rankings was highly significant (Gamma test: $G = 0.88$; $P \ll 0.001$). Consequently, only FA1 which presented a higher concordance between traits was considered in further analyses.

Relationships between genetic diversity and environmental features

Nevo *et al.* (1994a), using a stepwise model of multiple regression analysis, found that, among numerous environmental features, the number of rainy days per year (RD) correlated most closely with the indicators of genetic diversity. This was established on 12 populations from Israel. In our study, 14 populations from Israel and one from Egypt were considered and the relationships between environmental features and genetic diversity had to be reassessed. None of the correlation statistics obtained between all the genetic diversity indicators and all the environmental features was found to be significant (Table 4). Stepwise models of multiple regression analysis (not reported here) showed that for most of the genetic diversity indicators (P1%, P5%, H and HE), RD

Table 4 Pearson correlation between environmental and genetic variables.

	Environmental variables							
Genetic variables	ALT	TJ	TA	TM	TD	RN	RD	HU
Including EGY								
A	−0.07	0.12	0.06	0.17	−0.15	−0.21	−0.21	−0.15
P1%	−0.01	0.12	−0.01	0.12	−0.24	−0.22	−0.30	−0.16
P5%	0.02	0.17	0.03	0.12	−0.28	−0.29	−0.40	−0.09
H	0.02	0.15	0.02	0.08	−0.26	−0.27	−0.35	−0.34
HE	0.00	0.16	0.03	0.11	−0.27	−0.24	−0.36	−0.19
Excluding EGY								
A	−0.25	0.42	0.21	0.35	−0.52	−0.41	−0.43	0.05
P1%	−0.18	0.43	0.13	0.29	−0.64*	−0.42	−0.53	0.04
P5%	−0.06	0.32	0.10	0.20	−0.49	−0.39	−0.53	0.00
H	−0.14	0.47	0.18	0.24	−0.67**	−0.48	−0.60*	−0.18
HE	−0.15	0.46	0.17	0.26	−0.65*	−0.42	−0.59*	−0.02

*: $P < 0.05$; **: $P < 0.01$.

was the first factor included in the model, but in all these cases the multiple regression remained nonsignificant. The addition of the Egyptian population could have been responsible for the discrepancy between Nevo's results (Nevo *et al.*, 1994a) and ours. When the Egyptian population was excluded from the data set, the correlation statistics appeared to be significant between P1%, H and He indicators with RD and TD (Table 4). This analysis supported the assumption of Nevo *et al.* (1994a) that aridity and the genetic diversity were significantly correlated among Israeli localities. However, the inclusion of the Egyptian locality (EGY), for which the level of heterozygosity was lower than expected on the basis of the Israeli pattern, altered the relationships between aridity and genetic diversity.

Relationships of FA levels with environmental features

Gamma tests of rank correlation between environmental variables and Rj (Table 5) indicated that FA was positively correlated with altitude and seasonal temperature differences (ALT and TD) and negatively with the mean temperature in January (TJ) (Fig. 2). The discrepancy between the signs of the correlation was expected since ALT and TD were negatively correlated with TJ (ALT–TJ: $r = -0.90$**; TD–TJ: $r = -0.74$**).

The survey of the relationship between FA levels for each trait with all the environmental features showed that API is correlated with seven of the eight environmental features considered. Aditionally, Kruskall–Wallis tests performed on FA traits indicated that the level of FA displayed by the API trait and Rj were dependent on soil induration: the harder the soil, the higher the level of FA (Table 5). Although, the effects of environmental stresses on FA levels were observed, their origin could not be ascribed to climatic or soil features.

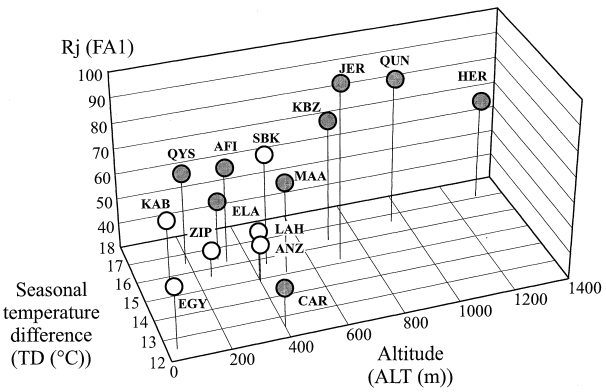

Fig. 2 Plots of synthetic levels of fluctuating asymmetry (Rj) for populations against altitude (ALT) and seasonal difference of temperature (TD). Closed circles: populations in hard soil; open circles: populations in light soil.

Table 5 (a) Pearson coefficients of correlation of genetic and environmental variables with FA indices (except for Rj: Gamma rank correlation); (b) Kruskall–Wallis H statistics for soil effect analyses on FA estimator.

	Fluctuating asymmetry estimators								
	r								G
	API	TI	LM1	WM1	LM2	WM2	LM3	WM3	Rj
(a)									
Genetic vairables									
A	−0.25	0.10	−0.27	−0.23	−0.14	−0.21	0.20	0.13	−0.21
P1%	−0.28	0.01	−0.24	−0.22	−0.09	−0.06	0.12	0.18	−0.09
P5%	−0.32	−0.28	0.04	−0.22	−0.11	0.19	0.26	0.24	0.07
H	−0.30	−0.24	0.02	−0.16	−0.17	0.26	0.12	0.08	−0.08
HE	−0.25	−0.16	0.01	−0.32	−0.14	0.15	0.11	0.15	−0.02
Environmental variables									
ALT	0.71**	0.05	0.65**	0.48	0.06	0.46	−0.13	−0.20	0.43*
TJ	−0.77***	−0.27	−0.40	−0.50	−0.20	−0.17	0.21	0.10	−0.47*
TA	−0.64**	−0.11	−0.50	−0.29	−0.22	−0.31	0.22	0.30	−0.31
TM	−0.72**	−0.07	−0.60*	−0.33	−0.09	−0.36	0.26	0.41	−0.17
TD	0.62*	0.38	0.08	0.56*	0.08	−0.10	−0.10	0.22	0.39*
RN	0.68**	0.28	0.31	0.08	0.06	0.05	−0.20	−0.43	0.11
RD	0.61*	0.36	0.10	0.13	0.08	−0.13	−0.12	−0.44	0.15
HU	0.06	−0.05	0.17	−0.43	0.06	0.01	0.16	−0.32	0.02
(b)									
Soil (1:Hard; 2:light)†	7.34**	0.68	0.12	1.68	2.34	2.00	0.68	0.02	5.03*

*: $P < 0.05$; **: $P < 0.01$; ***$P < 0.001$

Relationships between FA levels and heterozygosity

Testing the rank correlation between FA estimators and each of the genetic diversity estimators revealed no statistical significance considering either each FA trait or each Rj. Testing the relationships between each of the eight FA estimators and locus by locus heterozygosity, for the 25 polymorphic ones, led to 200 correlation coefficients. Applying one-tailed tests for the expected negative correlation between FA and heterozygosity led to as few as seven significant statistics, which could definitively be imputable to type 1 error. These results clearly supported the lack of statistical relationships between heterozygosity and FA levels in our case.

Discussion

Developmental stability and environmental features

Adaptive radiation within the chromosomal superspecies of *S. ehrenbergi* in Israel has been suggested to involve speciation in semiarid and arid climates by physiological adaptation (metabolism, kidney conservation of water) along with multiple morphological, ecological and behavioural adaptive syndromes to increasing aridity (Yahav *et al*., 1988, 1989; Nevo, 1991; Ganem & Nevo, 1996). Developmental stability estimated by the synthetic and relative level of FA, Rj, based on all tooth traits and used as a measure of habitat suitability in the mole rat populations, was not found to be related to the global climatic pattern of increasing aridity southwards. While aridity is considered as the major selective force acting on populations (and species) of the mole rat southwards, the lack of a relationship between developmental stability and aridity suggested that adaptation to this peculiar environmental trait is fairly achieved with no cost in terms of developmental stability.

In contrast, developmental stability appeared to be impaired by one or several covarying climatic features, i.e. altitude, mean January temperature and seasonal temperature difference and by soil induration. Although a global relationship between environment and FA was revealed among mole rat populations, it remained very difficult to weight the respective effects of altitude, temperature and soil on developmental stability, all these features being potentially stressful.

In a study on body size variation among 44 Israeli populations of *S. ehrenbergi*, altitude, rather than aridity, has been found to be the major determinant of body weight and length, verifying Bergmann's rule (Nevo *et al*., 1988). The increase of size under cooler environments is usually considered as a physiological adaptation limiting the loss of heat (Hoffmann & Parsons, 1991). The fact that higher altitudes and lower temperatures impair developmental stability in the mole rat could indicate a certain maladaption to cooler environments despite Bergmann's rule. It may also express the cost in high-altitude populations of the global adaptation to xericity for this Near-Eastern subterranean rodent: adaptation to aridity, mostly related to high temperatures in this region, might be physiologically costly in cooler environments.

Besides, the soil induration could also be considered as having a potential effect on developmental stability. Flynn *et al*. (1987) have shown that the thickness of incisor enamel, which differed among *S. ehrenbergi* chromosomal species, increased with soil induration, indicating that thickness could be advantageous for digging. Additionally, molar morphology has been shown to differ between populations according to soil type (Butler *et al*., 1993). If these dental traits are adaptive, soil induration may be considered as an efficient selective pressure able to induce environmental stresses.

We have noted that the morphological trait most correlated with environmental features is the anterio-posterior width of the incisor (API). Incisors in rodent are hypsodont, which means that they grew during the whole life of the animal. This contrasts with molars which, as in the house mouse (Bader, 1965), developed to their definitive size early in life. Further analyses would be required to assess the variability of asymmetry along the incisor, and its relationship with environmental changes during the life of the animal. This trait may provide an interesting marker more related to the environmental stresses undergone by animals during their life-time. Additionally, this may also provide some insight into the mechanisms which generate asymmetries (see for review Møller, 1996).

We should, however, point out that the cline of aridity is matched with several other biotic or abiotic clines, among which is the heterozygosity displayed by populations. An alternative hypothesis would be that aridity remains a stressful factor on *S. ehrenbergi* populations, but the related impairing effect on the stability of development, which is expected to increase southwards, could be complemented by increasing heterozygosity.

Developmental stability, genetic features and speciation events

The role of heterozygosity on FA has been the focus of extensive studies, and a negative relationship between these two features has been reported (Mitton & Grant, 1984; Clarke, 1993; Markow, 1995). Nevertheless, studies failing to show this relationship are not rare (Wooten & Smith, 1986; Patterson & Patton, 1990; Clarke, 1993; Yampolosky & Scheiner, 1994). The strongest evidence for such a dependence was certainly provided by intra-population studies showing that the most homozygous individuals for several allozymic loci displayed the higher levels of FA (Leary *et al*., 1983, 1984, 1992; Biémont, 1983). Mitton (1993) stressed that the association between heterozygosity and developmental homeostasis

may be attributable to a limited number of genes rather than to the entire genome and that the assessment of heterozygosity based on several allozymic markers may obscure the association between heterozygosity at a specific locus and developmental stability. In *S. ehrenbergi*, genetic indices have been established on subsamples (Nevo *et al*., 1994a) of those used for the present FA approach and were presumed to be reliable population indicators of the genetic diversity. Yet, the developmental stability in the mole rat was associated neither with the overall heterozygosity based on the 36 allozyme loci nor with the heterozygosity at each of the 25 polymorphic ones.

If we postulate that the constant level of developmental stability along with the sharp gradient of aridity supports the idea of an efficiency of adaptive strategies of populations to their local conditions of aridity, an increasing heterozygosity would then be expected to increase the level of developmental stability southwards despite the higher aridity stress. This absence of a detectable influence of heterozygosity on developmental stability in *S. ehrenbergi* populations may be then diversely explained. First, there may be no visible effect of heterozygosity on developmental stability as already reported in various studies (see references above). Second, the beneficial effect of heterozygosity on development could be counterbalanced by another effect impairing developmental stability. The southward increase in heterozygosity is accompanied by a number of environmental and genetic changes. For instance, a parasitological approach has shown that the number and co-occurence of coccidian species found in *S. ehrenbergi* increased in southern species ($2n = 58$ and 60) (Couch *et al*., 1993). Even though it may not be generalized (Alibert *et al*., 1994), parasites have been shown to decrease developmental stability (Møller, 1992; Parsons, 1992; Polak, 1993; Markow, 1995) and could be a relevant applicant to balance the benefit of heterozygosity on developmental stability. By contrast, if we postulate that aridity remains a stressful factor southwards, the alternative hypothesis for the lack of a relationship between FA with aridity and heterozygosity would simply express the complementing effects of these two features on developmental stability.

The developmental stability is believed to depend on both genomic coadaptation and heterozygosity (Clarke, 1993). It is thought that in a typical diploid organism, coadaptation refers to a relational balance between homologous chromosomes and an internal balance among genes within and among chromosomes (Mather, 1973 in Clarke *et al*., 1992). Chromosomal differences between mole rat species combine whole-arm Robertsonian changes (fusions but mainly fissions), pericentric inversions (Wahrman *et al*., 1985) and a considerable amount of chromosomal microchanges (Nevo, 1988). Nevo (1991) suggested that, on the basis of available molecular and organismal data, there was no evidence of a genetic revolution or of a major gene reorganization at each step of the radiation. However, each successively emerging species exhibited a new chromosomal organization of the genome through an increase in chromosomal number, which could modify both the pattern and the rate of recombination. Nevo *et al*. (1996) reported an increasing rate of recombination from $2n = 52$ species to $2n = 60$. Increased recombination rates may disrupt ancestral coadaptive gene complexes resulting in a 'recombinational load' (Charlesworth & Barton, 1996). Such an increase in recombination rate has been reported between the $2n = 52–54$ and the $2n = 58–60$ (Nevo *et al*., 1996). It could be then hypothesized that, in the *S. ehrenbergi* superspecies, the gradient in heterozygosity from ancestral to derived parallels that of an increase in recombination rates; their opposite effects would compensate along the gradients and maintain an equivalent level of developmental stability among the chromosomal species.

Obviously, further studies are required to examine this hypothesis. It implies that within each chromosomal species of the mole rat, a relationship between heterozygosity and developmental stability is expected. However, since trans-specific effects of environmental stresses, e.g. altitude, have been demonstrated in the present study, this approach has to be conducted at the intrapopulation level by grouping individuals according to their level of heterozygosity and thus implies that our sample sizes need to be considerably enlarged.

Conclusions

In order to analyse and interpret the level of developmental stability in natural populations, Clarke (1993) stressed the need to know the exact genetic structure and the evolutionary history of groups under examination. However, the more that is known, the higher is the number of potentially inextricable factors acting on developmental stability. Several authors, including Parsons (1988), Hoffman & Parsons (1991) and Clarke (1993), emphasized the complex relationships that exist between environmental and genetic stresses as well as their potentially cumulative effects on developmental stability. The assessment of natural habitat suitability using developmental stability among mole rat populations would lead us to conclude that lower altitude, lighter soil or hotter environments should not be considered as stressful, despite the fact that they can be drastically more arid. Aridity would then not appear as a stressful factor, even though it has been considered as the major selective pressure during the adaptive radiation of this superspecies towards xeric environments. Instead, the inclusion of potential genetic stresses in addition to environmental ones in this study of the developmental stability of natural populations of *S. ehrenbergi* clearly underlines the likelihood of complex complementary effects which hampers the

interpretation of habitat suitability in terms of adaptive strategies. Although hypotheses on such complementary effects may emerge from studies of natural populations, testing them definitely requires experimental analyses.

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