

Genetic divergence of heat production within and between the wild progenitors of wheat and barley: evolutionary and agronomical implications

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Summary. We compared and contrasted calorimetrically heat production in seedlings incubated at 5°C and 24°C using genotypes from cold and warm Israeli populations of the wild progenitors of barley (*Hordeum spontaneum*) and wheat (*Triticum dicoccoides*). The wild barley sample comprised 14 accessions, 7 from cold localities and 7 from warm localities. The wild emmer wheat sample consisted of 12 accessions, 6 from a cold locality, and 6 from a warm locality. Our results indicated that (1) heat production was significantly higher in the two wild cereals at 5°C than at 24°C; (2) interspecifically, wild barley generates significantly more heat than wild wheat at both 5°C and 24°C; (3) intraspecifically, wild barley from warm environments generates significantly more heat than wild barley from cold ones, at 24°C. We hypothesize that both the inter- and intraspecific differences in heat production evolved adaptively by natural selection in accordance with the niche-width genetic variation hypothesis. These differences presumably enhance biochemical processes, hence growth, thereby leading to the shorter annual cycle of barley compared to that of wheat, and may explain the wider range of the wild and cultivated gene pools of barley, as compared with those of wheat. We propose that a shortening of the growth period through utilizing heat production gene(s) is feasible by classical methods of breeding and/or modern biotechnology.

Key words: Genetic adaptation – Heat production – Thermogenesis – Wild barley – Wild wheat

Introduction

Temperature is an important environmental factor affecting the genetic evolution and geographical distributions of all organisms (Christiansen and Lewis 1982; Graham and Patterson 1982; Hochachka and Somero 1984; Cossins and Bowler 1987). Both low and high temperatures are important ecological factors limiting organismal evolution, distribution and survival (Graham and Patterson 1982). Ample comparative physiological evidence indicates that species (Hochachka and Somero 1984; Prosser 1986) and even conspecific populations (Watt 1985; Powers 1987) can adapt genetically to different temperature regimes and/or avoid extreme temperature stresses. However, the direct effect of these adaptations of fitness and the rapidity of their evolution is largely unknown.

The physiology of temperature effects on plants has been reviewed by McDaniel (1982). Every plant species is characterized by an optimum and range temperature for growth. The temperature response of the species is determined by a complex interplay of genetic, developmental and environmental factors. Plant evolution and biogeography as well as the growth period are determined by diverse factors amongst which temperature is distinct. Clearly, tropical plants possess markedly less cold tolerance than do arctic species (McDaniel 1982).

Remarkably, some plants have evolved heat production mechanisms that rival those observed in birds and mammals (McDaniel 1982; Meeuse and Raskin 1988; Raskin et al. 1989). Intense thermogenesis has been observed in the flowers and inflorescences of plants belonging to at least six families of angiosperms (Raskin et al. 1987; 1989). The rationale for heat production in the growing parts of plants may be as suggested by McDaniel (1982, p 29; see discussion).

In all of the cases studied plant thermogenesis was associated with an increase in the alternative or cyanide-insensitive respiratory electron transport system, a system that is unique to the mitochondria of plants, fungi and some protists (Siedow and Berthold 1986; Meeuse and Raskin 1988).

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Table 1. Ecogeographical background and thermogenesis in the wild cereals, *Hordeum spontaneum* and *Triticum dicoccoides*, in Israel, at 5°C and 24°C

Ecogeographic background

Number	Population	Ln ^a	Lt ^a	Al ^a	Tm ^b	Ta ^b	Tj ^b
1	Mt. Hermon	35.73	33.30	1300	11	21	3
9	Mt. Meron	35.40	33.05	1150	14	22	6
18	Gitit	35.40	32.10	300	21	29	13
20	Sede Boqer	34.78	30.87	450	19	29	12
22	Mehola	35.48	32.13	-150	23	31	14

^a Geographical Symbols: Ln, longitude (in decimals); Lt, latitude (in decimals); Al, altitude (m)

^b Temperature Symbols: Tm, mean annual temperature (°C); Ta, mean August temperature (°C); Tj, mean January temperature (°C)

Thermogenesis

Number	Population and plant #	n	Thermogenesis (micro-Watts/g dry weight)		5 – 24°C difference	
			24°C + SE	5°C + SE	Difference	% of 24°C
Wild barley						
<i>Cold populations</i>						
1	Mt. Hermon	2				
	01 34		8 769.7 + 524.3	12 053.3 + 550.6	3 283.6	37.4%
	01 46		8 964.3 + 513.9	13 294.5 + 2 575.2	4 330.2	48.3%
9	Mt. Meron	5				
	09 42		8 474.8 + 739.7	10 171.5 + 523.0	1 696.7	20.0%
	09 18		8 256.1 + 2 187.7	10 256.3 + 1 359.5	2 000.2	24.2%
	09 53		8 571.8 + 75.0	11 625.0 + 986.3	3 053.2	35.6%
	09 47		8 719.3 + 576.4	12 914.8 + 720.3	4 195.5	48.1%
	09 36		8 478.2 + 842.6	14 680.3 + 1 713.2	6 202.1	73.2%
Mean of cold			8 604.9 + 88.0	12 142.2 + 618.8	3 537.3	41.0%
<i>Warm populations</i>						
20	Sede Boqer	2				
	20 44		9 355.9 + 719.7	12 664.1 + 826.6	3 308.2	35.4%
	20 37		9 544.8 + 489.0	13 416.9 + 194.5	3 872.1	40.6%
22	Mehola	5				
	22 15		10 232.3 + 108.7	11 718.0 + 1 874.2	1 485.7	14.5%
	22 04		9 340.1 + 1 150.6	12 783.7 + 1 552.8	3 443.6	36.9%
	22 16		10 403.3 + 742.0	12 091.1 + 1 582.8	1 687.8	16.2%
	22 48		9 929.8 + 368.7	11 808.6 + 1 000.8	1 878.8	18.9%
	22 35		9 335.6 + 1 448.8	11 857.0 + 761.6	2 521.4	27.0%
Mean of warm			9 734.5 + 170.9	12 334.2 + 240.3	2 599.7	27.1%
Significance of cold-warm populations difference						
By Mann-Whitney tests			0.0006	ns	ns	0.097
By ANOVA			0.0001	ns	ns	ns
Total n			14			

This alternative system of respiration is also a major source of heat production in non-overtly thermogenic plants, which generate much less heat than thermogenic plants (Ordentlich et al. 1991). We tested the thermogenesis potentials of the wild progenitors of all cultivated barleys (*Hordeum spontaneum*) and cultivated wheats (*Triticum dicoccoides*). Both wild progenitors have been extensively studied as rich genetic resources for barley (Nevo et al., 1979, 1986; Nevo 1992) and wheat (Nevo and Beiles 1989) improvement.

In the investigation presented here we examined the differences in heat production in wild barley and wheat with emphasis on interspecific and intraspecific variations resulting from different growing environments. The results suggest an explanation for the differential distribution of wheat and barley.

Table 1. (continued)

Number	Population and plant #	n	Thermogenesis (micro-Watts/g dry weight)				5 – 24°C difference	
			24°C	+ SE	5°C	+ SE	Difference	% of 24°C
Wild wheat								
<i>Cold population</i>								
1	Mt. Hermon	6						
	01 01		8 213.7 +	167.0	9 443.1 +	436.0	1 229.4	15.0%
	01 03		6 014.0 +	457.0	9 568.8 +	936.0	3 554.8	59.1%
	01 39		8 126.8 +	502.0	12 559.3 +	1 203.1	4 432.5	54.5%
	01 56		7 553.5 +	411.9	9 077.8 +	657.7	1 524.3	20.2%
	01 52		9 317.6 +	686.2	9 837.8 +	268.6	520.2	5.6%
	01 42		4 806.0 +	412.3	8 318.8 +	772.7	3 512.8	73.1%
Mean			7 339.9 +	671.6	9 800.9 +	591.9	2 461.0	37.9%
<i>Warm population</i>								
18	Gitit	6						
	18 10		5 866.9 +	215.9	9 554.2 +	1 036.3	3 687.3	62.8%
	18 61		6 159.6 +	616.5	11 607.6 +	973.2	5 548.0	88.4%
	18 03		7 579.0 +	772.3	7 024.1 +	229.0	– 554.9	– 7.3%
	18 29		6 993.5 +	311.4	9 676.0 +	404.4	2 682.5	38.4%
	18 48		7 261.9 +	636.5	9 577.6 +	870.7	2 315.7	31.9%
	18 01		6 774.8 +	212.0	8 108.8 +	107.0	1 334.0	19.7%
Mean			6 772.6 +	266.7	9 258.1 +	637.6	2 485.4	39.0%
Significance of cold-warm populations difference.								
By Mann-Whitney tests			ns		ns		ns	ns
By ANOVA			ns		ns		ns	ns
Total		12						
Significance of wild wheat-barley comparison								
By Mann-Whitney tests			0.0001		0.0001		ns	ns

Materials and methods

Plant material

The analysis was conducted on wild barley (*Hordeum spontaneum*) from four natural populations, two cold (Mt. Hermon and Mt. Meron) and two warm (Mehola and Sede Boger), and on wild emmer wheat (*Triticum dicoccoides*) from two populations, one cold (Mt. Hermon) and one warm (Gitit). The ecogeographical background of the two wild cereal populations appear in Table 1 and is also described in detail in Nevo et al. (1979) and Nevo and Beiles (1989) for wild barley and wild emmer wheat, respectively. The seedlings were grown in an environmentally controlled growth chamber at 24°C and 25% RH and exposed to a 16-h photoperiod (600 mmol m⁻² s⁻¹ provided by a combination of incandescent and cool white fluorescent lights). Prior to the experimental treatments 4-week-old plants were preincubated for 24 h at 24°C in continuous light (300 mmol m⁻² s⁻¹). During the experiments plants were incubated at 5°C or 24°C in continuous light (300 mol m⁻² s⁻¹) for 10 h.

Calorimetric measurements

Calorimetric measurements were made with a Hart scientific 7707 DESC (differential scanning microcalorimeter) (Pleasant Grove, Utah). Three simultaneous measurements were made in 1-cm³ ampules (10 mm diameter). Three leaf samples (10 mm

length) were placed in each calorimetric ampule containing 50 ml H₂O. All calorimetric measurements were performed at 20°C after 1 h equilibration using the isothermal operation mode. All experiments included at least three replicates and were repeated at least 3 times with similar results.

Statistical analysis

We used the SPSSx (1986) statistical package. The mean differences in heat production were tested both by the parametric ANOVA and by the non-parametric Mann-Whitney or sign test.

Results

The results appear in Table 1 and Fig. 1. Thermogenesis was measured in all 26 genotypes of the two wild cereal species.

Heat production by wild emmer wheat and barley

Thermogenesis at low and high temperatures. Heat production by plants (or seedlings) exposed to 5°C for 10 h was significantly higher than by those exposed to 24°C,

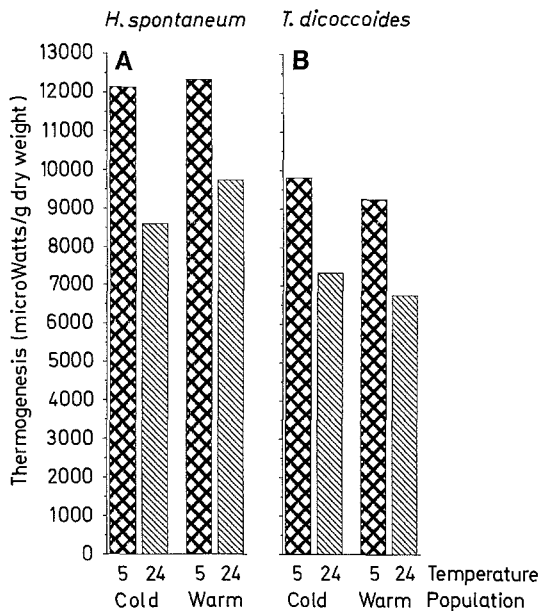


Fig. 1A, B. Heat production in wild barley (*H. spontaneum*) (A) and wild emmer wheat (*Triticum dicoccoides*) (B) from cold and warm populations incubated at 5° and 24°C

and was found in 25 out of the 26 genotypes ($P < 0.000001$, sign test).

The two wild cereals are genetically polymorphic both inter- and intraspecifically for heat production.

Interspecifically, wild barley generated significantly ($P < 0.0001$) more heat than wild emmer wheat after incubation at both 5°C and 24°C. Intraspecifically, wild barley from warm environments (Mehola and Sede Boqer) grown at 24°C generated significantly ($P < 0.001$) more heat than wild barley from cold environments grown at 24°C (Mts. Meron and Hermon), while those that were grown at 5°C exhibited just a small non-significant difference in the same direction. Wild emmer wheat showed intraspecifically a non-significant opposite trend to that of wild barley, both at 5°C and 24°C. The Mt. Hermon cold population had a higher level of heat production than that of the warm Gitit population. In addition, at 24°C the Hermon population showed a wider polymorphism than Gitit.

Discussion

Alternative respiration or the cyanide resistance pathway is common in all plants (Sidow and Berthold 1986) and dissipates most of the chemical energy of mitochondrial respiratory substances as a quantifiable heat that can be detected by calorimetry (Ordentlich et al. 1991). The "energy overflow" hypothesis (Palmer 1976; Lambers 1980) suggests that the alternative pathway dissipates excess reducing power in situations in which energy and ATP

are not limiting or when the cytochrome C (Cyt) pathway is saturated. The function of this seemingly bioenergetically wasteful process is explicable in thermogenic species, in which alternative pathway-evolved heat is used either to attract insect pollinators (Meeuse and Raskin 1988) and/or to enhance growth (McDaniel 1982, p 29).

Plants exposed to a chilling temperature responded by production of heat that could be blocked by alternative respiration pathway inhibitors, but not by cytochrome c pathway inhibitors (Ordentlich et al. 1991). If the within-cell temperature is elevated by thermogenesis then it can be expected that the biochemical processes of germination or growth will be accelerated. This may be critical under ecologically marginal conditions, because the potential enhancement of plant growth may enable the shortening of the life cycle, hence be advantageous either in warm or cold deserts. Thus, the apparent wastage of a small part of the photosynthetic products for thermogenesis may result, through a positive feedback mechanism, in a higher photosynthetic yield that will enhance growth.

The intraspecific genetic polymorphism in heat production between wild barley populations may be evolutionarily important. It presumably enables the steppic (Mehola) and desert (Sede Boqer) populations to complete their annual cycle about 1–2 months earlier than the cold mountain populations (Mts. Meron and Hermon). Consequently, the early ripening of desert populations is adaptive in that it enables an evasion of the summer heat, severe drought and desiccation that characterizes early summer in the desert. The significantly higher heat production in the cold period (5°) also supports the adaptive explanation: it may enhance growth when it is limited by the ambient temperature. Increased heat production may be responsible for the shortening of the annual cycle.

The interspecific genetic polymorphism in heat production between the two species of wild cereals is distinct. Heat production is always higher in barley than in wheat. This differential may largely explain the dramatic differences in the geographical distribution of both the wild and cultivated gene pools of the two cereals. The growing period of barley is shorter than that of wheat, which may be a result of higher heat production. Therefore, both wild and cultivated barley are by far more extensive in distribution than wild and cultivated wheats, respectively. Wild barley penetrates into the desert and extends deep into Central Asia, reaching China. Cultivated barley reaches its upper limit of cultivation in high mountains and may be grown in desert oases. Barley is a cool-season crop; it can tolerate high temperatures if the humidity is low, but is not suited to warm, humid climates and is only grown in the tropics in cool highlands, as in Mexico, the Andes, and East Africa (Simmonds 1976). In contrast, wild emmer wheat is limited to the Near East

Fertile Crescent, primarily northern Israel (Nevo and Beiles 1989). The intraspecific non-significant opposite trend found in wild wheat as compared to wild barley (Table 1) may derive from the narrower ecological (climatic) niche of the former. The distribution of cultivated wheat is globally extensive, but again more restricted than that of barley (Simmonds 1976). Cultivated barley extends farther north into the Arctic region than wheat in both North America and Europe.

We suggest that the significant genetic divergence in heat production between wheat and barley evolved adaptively by natural selection and contributes substantially to the shorter growth period and to the broader ecological niche of barley as compared with wheat in both the wild and cultivated gene pools. This endows barley with a better colonizing and competitive ability as well as broader global range of cultivation than wheat (Simmonds 1976).

Remarkably, this pattern is also exhibited by the level of gene diversity based on allozyme diversity. The genic diversity of wild barley is significantly higher ($H_e = 0.108$, range 0.0–0.194; Nevo et al. 1986) than that of wild emmer wheat ($H_e = 0.059$, range 0.002–0.119; Nevo and Beiles 1989), which is in accordance with the niche-width variation hypothesis (Van Valen 1965) that predicts a positive correlation between genetical and ecological diversities. This substantial interspecific difference in heat production between barley and wheat, as well as the intraspecific variation, is largely genetic because it was found under standardized greenhouse conditions. Therefore, the intraspecific and even the interspecific genetic differences can be used agronomically by breeding for shortening of the growing season either by classical (Marshall 1987) or modern (Schell 1987) biotechnology.

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