

Physiological-genetic dissection of drought resistance in wild emmer wheat

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ABSTRACT

Wild emmer wheat (*Triticum turgidum* ssp. *dicoccoides* (Körn.) Thell.), the allo-tetraploid (BBAA) progenitor of cultivated wheats, offers a valuable source of allelic diversity for various economically important traits, including drought resistance. A total of 160 wild emmer accessions, consisting of 25 populations, and three control durum wheat cultivars were examined under two irrigation regimes, well-watered control (~650 mm) and water-limited (~250 mm). Principal component analysis, supported by photosynthetic rate measurements, revealed a variety of drought adaptive strategies among the wild accessions. A wide phenotypic variation was found both between and within the wild emmer populations in most morpho-physiological traits, with a considerable advantage in drought resistance over cultivated genotypes. Microsatellite markers revealed a wide allelic diversity between and within the wild emmer populations, confirming the patterns of phenotypic variation. The greatest drought resistance capacity corresponded with the highest allelic diversity and found in populations from intermediate aridity level. Physiological responses to drought were further dissected by quantitative trait loci (QTLs) mapping of yield and drought related traits under contrasting irrigation regimes, using 152 F₆ recombinant inbred lines derived from a cross between durum wheat and wild emmer wheat from drought-prone environment. A total of 102 QTLs were mapped for 10 productivity and morpho-physiological traits. Several QTLs exhibited GxE interaction and accounted for productivity and related physiological traits under either the well watered or water-limited conditions. The identified genetic resources and QTLs detected, shed new light on drought adaptive complexes in wheat and expected to facilitate the improvement of drought resistance in elite wheat cultivars.

INTRODUCTION

Drought is the main environmental factor limiting wheat productivity worldwide. Moreover, climate-change scenarios predict an increased aridity in many regions of the world. The development of novel cultivars with more efficient water-use and greater drought resistance capacity is considered a sustainable and economically viable solution to this problem. The implementation of this solution requires wide explorations of potential genetic resources and in depth understanding of the

ecophysiological and genetic adaptive mechanisms and responses to water deficiency.

The genetic diversity of crop plants has been tremendously eroded as compared with their wild progenitors throughout plant domestication and subsequent breeding. As a result, modern crop plants have become more vulnerable to environmental stresses. Therefore, a major objective of modern breeding is to identify, in the wild ancestors of crop plants, valuable "left behind" alleles and introduce them into cultivated crops (Aaronsohn 1910; Tanksley & McCouch 1997).

Wild emmer wheat [*Triticum turgidum* ssp. *dicoccoides* (Körn.) Thell.] is the allo-tetraploid (2n=4x=28; genome BBAA) progenitor of cultivated wheats. It is fully compatible with the tetraploid (2n=4x=28; BBAA) durum wheat [*T. turgidum* ssp. *durum* (Desf.) MacKey] and can be crossed with the hexaploid (2n=6x=42; BBAADD) bread wheat (*T. aestivum* L.) (Feldman & Sears 1981). Wild emmer is distributed throughout the Near Eastern 'Fertile Crescent', across a variety of ecological conditions. Hence, wild emmer gene pool offers a rich allelic repertoire required for improvement of agronomically important traits. However, until recently very little have been published on drought responses in wild emmer.

In this paper, we summarize a long term study aimed to establish the basis for utilizing the wild emmer gene pool for improving drought resistance of the domesticated wheat species. The specific objectives of this study were:

- (i) Characterization of the genetic diversity for drought resistance in wild emmer wheat germplasm and its underlying physiological mechanisms.
- (ii) Characterization of allelic variation between wild emmer accessions at the DNA level and its association with their eco-geographical distribution.
- (iii) Identification of genomic regions associated with wheat response to water-deficit.

GENETIC DIVERSITY FOR DROUGHT RESISTANCE AND ITS UNDERLYING PHYSIOLOGICAL MECHANISMS

A total of 160 wild emmer accessions, consisting of 25 populations collected across natural aridity gradient in Israel, were examined during three years in rain protected field plot under two irrigation regimes, well-watered control (~650 mm) and water-limited (~250 mm). Three durum wheat cultivars were also examined

as controls. Wide genetic diversity was found, both between and within wild emmer wheat populations (Peleg et al. 2005). About 80% out of 110 wild emmer accessions studied in one trial produced, under water limited field conditions, greater total dry matter (DM) than domesticated durum lines (Fig. 1B). Selected lines, tested in larger plots in a subsequent year, further confirmed these results. Moreover, although wild emmer was never subjected to direct selection for yield or harvest index, about 30% out of 110 wild emmer accessions produced greater spike DM (representing reproductive biomass) under water deficiency, than the cultivated durum lines (Fig. 1A). A considerable number of wild emmer accessions exhibited higher stability (low drought susceptibility indices; Fischer and Maurer 1978) for both spike DM and total DM (Fig. 1) as compared with durum wheat cultivars, reflecting greater drought adaptations in wild emmer accessions. Seven out of the 25 wild emmer wheat populations examined (namely: Achihood, Yehudiyya, Gitit, Givat Koach, Kokhav Hayarden, Ma'ale Merav and Mt. Gilbboa) combined high productivity under water-limited conditions with higher stability (Peleg et al. 2005), thus offering the greatest potential for wheat improvement.

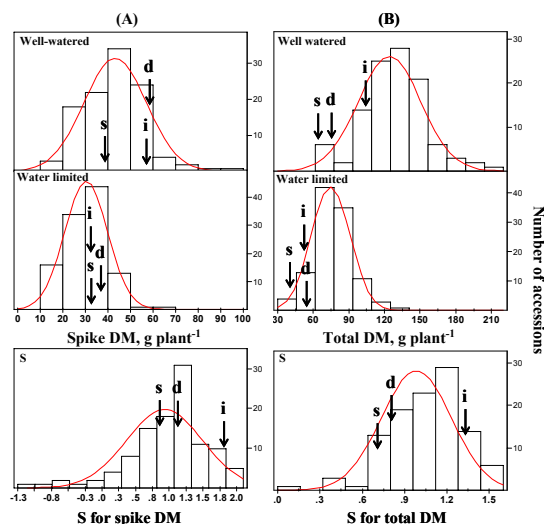


Figure 1. Frequency distribution of 110 wild emmer wheat accessions for the spike dry matter (A) and total dry matter (B) under well-watered and water-limited irrigation regimes, as well as in terms of drought susceptibility index (S). Arrows indicate the values of three durum wheat cultivars (d=580D, i=Inbar and s=Svevo). Data adapted from Peleg et al. (2005).

Under Mediterranean conditions, water deficit and high temperature during the grain-filling period (*i.e.*, terminal drought) are very common, causing dramatic reductions in crop yields. Terminal drought escape through early reproduction has been a very successful breeding strategy for Mediterranean environments. While under optimal growing conditions, late flowering leads to a longer vegetative growth period that promotes the accumulation and allocation of resources to seed production, early flowering (drought escape) improves

fitness in environments with a short or unpredictable growing season. In wild emmer wheat, spike DM and total DM were found associated with earliness, however, the greatest productivity was obtained in accession with mid-early (75-85 days from planting to heading) and not the earliest (65-75 days) heading date. Moreover, cultivated durum lines exhibited earlier heading than wild emmer accessions, but yet the latter exhibited greater drought resistance capacity. These results suggest that under natural selection, earliness is certainly useful but not the only component of drought adaptation. Wild emmer wheat may have evolved greater resistance to terminal drought, enabling better physiological status during grain filling period.

Water-use efficiency (WUE, the ratio between yield production and water consumption) is an important character determining crop performance under limited water availability. Stable carbon isotope ratio ($^{13}\text{C} / ^{12}\text{C}$, expressed in differential notation as $\delta^{13}\text{C}$) in plant tissues is a well established indirect measure of WUE. Under water-limited environment, wild emmer exhibited a wide genetic diversity for $\delta^{13}\text{C}$ as well as an advantage over durum wheat cultivars (Peleg et al. 2005). Moreover, most wild emmer accessions showed a considerably higher phenotypic plasticity for $\delta^{13}\text{C}$ (increased WUE under water deficiency) as compared with the cultivated durum controls. High WUE (and $\delta^{13}\text{C}$) can be achieved either through increased photosynthetic rate or reduced stomatal conductance, but only the former strategy would increase productivity. In agreement with this, principal component analysis revealed that in water-stressed wild emmer wheat high $\delta^{13}\text{C}$ was associated with either high or low productivity (Peleg et al. 2008, in press). The contrasting responses of WUE and productivity were supported by photosynthetic rate measurements of selected accession. These findings suggest that adaptive mechanisms for high productivity and high WUE can be improved in cultivated wheat by introgression from its wild relatives.

ALLELIC DIVERSITY ASSOCIATED WITH NATURAL ARIDITY GRADIENT

The 25 wild emmer wheat populations were genotyped with 54 microsatellite markers distributed throughout the 14 chromosomes of tetraploid wheat. Phylogenetic tree constructed based on the microsatellite allelic diversity revealed six major clusters (Peleg et al. 2008a). Several clusters were comprised of populations from ecologically similar but geographically remote habitats. Furthermore, genetic distances between populations were independent of the geographical distances. These results suggest that ecological determinants play an important role in shaping the genetic structure of wild emmer wheat.

Analysis of molecular variance revealed a wide allelic diversity, both between (56%) and within (44%) wild emmer wheat populations, in agreement with the phenotypic diversity. The within population allelic diversity exhibited a unimodal association with several environmental factors, indicating that the highest

diversity have evolved in habitats with intermediate environmental stress (e.g., rainfall 350-550 mm/year) (Peleg et al. 2008a). These habitats are subjected to a wide inter- and intra-annual environmental variability that impose fluctuating selection pressure leading to high genetic diversity. Moreover, six out of the seven populations that combined high productivity and stability across water availability treatments (see above) were originated from such intermediate environments. Surprisingly, wild emmer accessions from the most xeric habitats (<300mm rainfall per annum) exhibited relatively low drought resistance capacity as well as low microsatellite markers allelic diversity. It is hypothesized that these populations exploit a drought escape strategy either on an annual (producing in rainy years) or spatial (growing in wet micro niches) scale and thus being able to survive in such harsh environments.

GENOMIC DISSECTION OF DROUGHT RESPONSES

One accession (G18-16) representing the selected populations (see above) was crossed with domesticated durum cultivar (Langdon) to produce a mapping population. One-hundred and fifty two F₆ recombinant inbred lines (RILs) were analysed with a total of 690 loci, including 197 microsatellite and 493 DArT markers. A stabilized (skeleton) map consisting of 307 markers was established with a total length of 2317 cM and average distance of 7.5 cM between adjacent markers (Peleg et al. 2008b).

The RIL population was evaluated in the field under two contrasting water availabilities conditions for productivity and drought adaptive traits. Wide genetic variation and transgressive segregation were found among RILs for productivity and drought related morpho-physiological traits. A total of 102 quantitative trait loci (QTLs) were mapped for 10 traits reflecting plant productivity, phenology and physiology, with LOD score range of 3.0-35.4 (Peleg et al. 2008 submitted). A number of QTLs showed environmental specificity, accounting for productivity and related traits under water-limited (19 QTLs) or well-watered (15 QTLs) conditions, as well as drought-susceptibility index (21 QTLs). Major genomic regions controlling productivity and related traits were identified on chromosomes 2B, 4A, 5A and 7B. QTLs for productivity were associated with QTLs for drought-adaptive traits suggesting the involvement of several strategies in wheat adaptation to drought-stress.

FACING THE FUTURE: WILD EMMER AS POTENTIAL SOURCE FOR WHEAT IMPROVEMENTS

During a long evolutionary history across a range of environmental conditions wild emmer has accumulated a wealth of genetic diversity. Our comprehensive survey of wild emmer populations from across aridity gradient in Israel revealed a wide phenotypic and allelic diversity for drought response (Peleg et al. 2005, 2008a),

demonstrating the potential of the wild gene pool for wheat improvement. The greatest allelic diversity and drought resistance capacity found in wild emmer populations from intermediate-stress habitats suggesting that high priority should be given to conservation of these populations. The large proportion of the genetic variation residing within populations, as well as the various drought resistance strategies identified within the wild emmer gene pool, call for further exploration of promising population and for studying their adaptive mechanisms. Genetic dissection of drought related traits, through QTL analysis sheds light on drought adaptive mechanisms (Peleg et al. 200#, submitted). These results exemplify the unique opportunities to exploit favorable alleles that were excluded from the domesticated gene pool and may serve as a start point for introgression of promising QTLs into elite cultivated materials via marker-assisted selection.

ACKNOWLEDGEMENTS

This work was supported by The State of Israel, Ministry of Science, Plant Genetic Resources Program, grant #01-21-00048, The Israel Science Foundation grant #1089/04.

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