Wheat Wild Relatives – A 25 Year Program of *ex-situ* Conservation

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Introduction

Recent wide-scale urbanization and road construction are of immediate danger to many populations of wild-growing relatives of crop plants in which Israel is rich, among them relatives of wheat. In most parts of the tiny densely populated Israel the ideal solution of *in-situ* conservation cannot be applied.

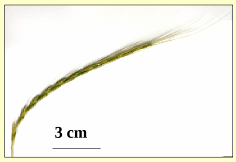
A long-term project of *ex-situ* conservation of wild emmer and of five diploid species of *Aegilops*, belonging to section Sitopsis was launched in 1980.

Table 1. Taxa collected for conservation

	Common name	Genome
Triticum turgidum L. subsp. *dicoccoides* (Körn. ex Aschers. & Graebn.) Thell	Wild Emmer	AABB
Aegilops bicornis (Forssk.) Jaub & Spach		S^bS^b
Ae. *longissima* Schweinf.& Muschl. emend. Eig	Slender Goatgrass	S^lS^l
Ae. *searsii* Feldman & Kislev ex Hammer	Sear's Goatgrass	S^sS^s
Ae. *sharonensis* Eig	Sharon Goatgrass	S^lS^l
Ae. *speltoides* Tausch var *speltoides*	Truncate Goatgrass	SS
Ae. *speltoides* Tausch var. *ligustica* (Savign.) Fiori	Truncate Goatgrass	SS

The goal of this project was to collect and store as large a number of genotypes as possible in each of these species. So far the collections have been used for evaluation of disease responses, salt tolerance and of molecular variation.

Figure 1.. A dense stand (yellow in foreground) of *Ae. sharonensis.* About 1 km behind it are the three chimneys of a coal-fuelled power station (the largest in Israel), forewarning of a sad future for this population.



Ae. sharonensis

Methods

Sampling – With the intention of garnering a maximum of genetic variation, collections of each species were made in diverse habitats and in different geographical regions. Usually, single-plant samples of one spike were taken at distances of about 5 m between plants along linear transects. In large populations, several parallel and/or perpendicular transect were plotted. In many cases the same population was sampled during a period of several years.

Recording - Each spike was recorded as a separate accession within the population. The site of each population and the lay-out of the transects were recorded using map coordinates or, more recently, GPS instruments. Notes were taken on soil type, annual rainfall, and foliar diseases.

Propagation – A S_1 generation of each accession in these species was raised from a single seed in each case in a nethouse in Tel Aviv University. Later generations progenies were produced on field plots for diverse tests.

Evaluations

Disease response – Seed of the original spike, as well as second-generation progenies were grown in temperature controlled greenhouse (20°C) for tests of response to artificial inoculation of seedlings with the foliar diseases: leaf rust, stem rust, stripe rust, and powdery mildew of wheat.

Molecular variation – Extracts from second generation seedling leaves were analyzed using RFLP, AFLP and microsatellite markers.

Salt tolerance – Progenies of advanced generations were used for this test.

Seed storage – The original seed of all accessions, as well as seed of progeny generations stored in paper bags in dry storage rooms at 5-10°C and 35% RH in the Lieberman Okinow Germplasm Bank at the Institute for Cereal Crops Improvement, Tel Aviv University. At present there are 13,476 accessions of the six species in the germplasm bank.

Genetic variation (molecular markers)

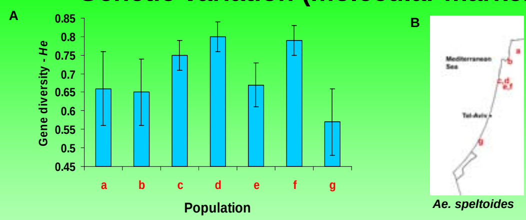


Figure 2. Genetic diversity, *He* of 7 populations of *Ae. speltoides* based on 8 micorsatellite markers. Highest *He* values were found in 3 populations (c,d,f) growing in bare patches in natural scrubland.

The lowest genetic diversity was found in an open field in Brachia (g) the southernmost world population of this species. Within populations diversity (86%), was much higher then among populations (14%).

The origin of the microsatellites is wheat. They where highly polymorphic (up to 36 alleles per locus). The diversity was analyzed under the infinite allele model.

A. Genetic diversity-*He* in 7 populations of *Ae. speltoides.*

B. Geographical location of the 7 populations on Israeli coastal plain schematic map.

Variation in host resistance

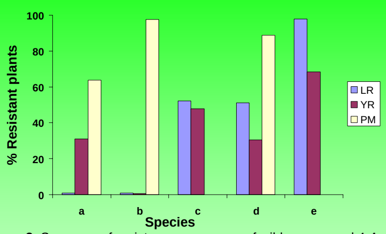


Figure 3. Summary of resistance responses of wild emmer and 4 *Aegilops* species to artificial seedling inoculations with wheat leaf rust (LR), wheat stripe rust (YR) and powdery mildew (PM) of wheat.

a = wild emmer
b = *Ae. bicornis*
c = *Ae. longissima*
d = *Ae. sharonensis*
e = *Ae. speltoides*

For *Ae. longissima* and *Ae. speltoides* no data of resistance to powdery mildew are shown.

It is noteworthy that *Ae. bicornis* (b) a desert species, shows almost total resistance to powdery mildew and no resistance to the rusts. Highest resistance to wheat leaf rust is in *Ae. speltoides (e)* while wild emmer (a), was highly susceptible to this rust.

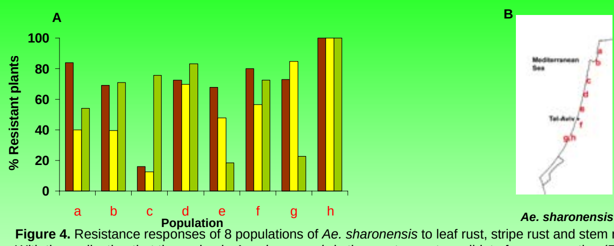


Figure 4. Resistance responses of 8 populations of *Ae. sharonensis* to leaf rust, stripe rust and stem rust of wheat.

With the realization that the endemic *Ae. sharonensis* is the most urgent candidate for conservation (Fig.1), greater emphasis has been placed on seed collections of this species in recent years.

A comparison of resistance responses to rust fungi of wheat – showed variation among populations of this species.

Two very different resistance pictures were obtained at the southernmost populations shown here (g and h) which grow in a close vicinity in the same sandy habitat.

A. Response of 8 populations of *Ae. sharonensis* (a-h) to seedling artificial inoculation with wheat leaf rust ■, wheat stripe rust ■ and wheat stem rust ■ in temperature controlled greenhouse.

B. Geographical location of the 8 populations on Israeli coastal plain schematic map.

Ammiad *in-situ* studies

For over two decades (since 1983) populations of wild emmer have been monitored and subjected to *in-situ* management at Ammiad, in Eastern Galilee. On-site studies have centered on demography, response to grazing regimes, and incidence of foliar diseases. Progenies from the site (taken annually from permanent sampling points), have been analyzed for spatial and temporal genetic variation in allozymes, subunits of storage proteins, AFLP and RFLP patterns, response to fungal pathogens, and variation in phenotypic traits. These progenies form part of our *ex-situ* stored collections.

Figure 5. A mixed population of wild emmer with wild barley (*Hordeum spontaneum*) at the Ammiad *in-situ* study site.

Parasitic variation – the mirror image

The great variation of the host has a mirror image – variation in the obligate parasites which evolved together with their hosts. Our research on rusts of wild relatives of wheat has led to a better understanding of their biology including discovery of new rust forms (Ben Yehuda *et al.* 2004).

Figure 6. *Puccinia triticina* f.sp. *speltoides, life cycle.*
Left - uredinial sori on leaves of *Ae. speltoides.*
Right - pycnial and aecial clusters on leaves of the alternate host – *Thalictrum speciosissimum* L.

Conclusions

Only part of the populations that we have sampled in the course of 25 years still exists today. The lost populations will be perpetuated by *ex-situ* conservation, if only partially.

Our studies point to considerable variation in the wild populations of each of the six species, including factors of disease resistance, (Anikster *et al* 2005 a,b)

All collections are held in abeyance for use of evolutionary research and most importantly, use by geneticists and breeders, now or in the future.

References:

1. Ben Yehuda, P., Eilam, T., Manisterski, J., Shimoni, A., and Anikster, Y. 2004. Leaf rust on *Aegilops speltoides* caused by a new forma specialis of *Puccinia triticina.* Phytopathology 94:94-101.
2. Anikster, Y., Manisterski, J., Long, D.L., and Leonard, K.J. 2005 a. Resistance to leaf rust, stripe rust and stem rust in *Aegilops* spp. In Israel. Plant Dis. 89: 303-308.
3. Anikster, Y., Manisterski, J., Long, D. L., and Leonard, K. J. 2005 b. Leaf rusts and stem rust resistance in *Triticum dicoccoides* populations in Israel. Plant Dis. 89: 55-62.