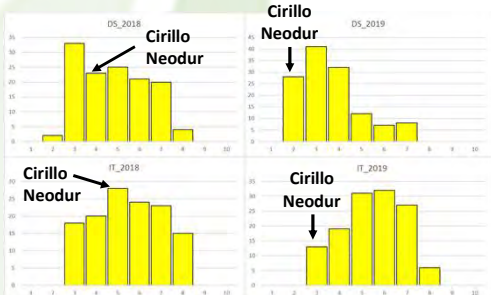


Durum wheat (*Triticum turgidum* L. var. *durum*) is a very important crop in the Mediterranean basin. Intense breeding activities are carried out in order to improve its productivity, quality and resistance, and new genomic tools are essential to speed up the breeding progress. The recent emergence of new widely virulent and aggressive strains of rusts (particularly stripe rust) is threatening durum wheat production, especially under the trend of higher temperature and humidity. A big effort has been undertaken to explore the genetic variability for resistance to these fungal pathogens and discovering novel resistance genes in both wild and elite gene pools to capitalize the new resistance sources in pre-breeding and breeding programs.

## QTL analysis in the Cirillo x Neodur RIL population

A segregating population of 150 recombinant inbred lines (RILs), derived from the cross between the durum wheat cv. Cirillo and Neodur, was evaluated for response to stripe rust in field under natural infection conditions for 2 years (3 biological replications) in terms of disease severity and infection type (scale 1-9). Heading date was also evaluated in terms of days from the 1<sup>st</sup> of April to investigate if growth stage could influence the plant response to the infection, and no influence was identified. Phenotypic data and the genetic map (400 markers per 1,989.6 cM) were integrated to run a Composite Interval Mapping QTL analysis with the QGene software.



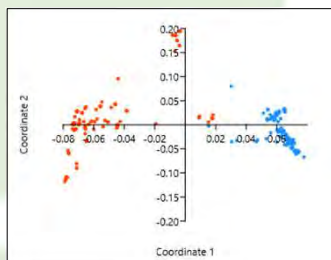
In both years of evaluation, Cirillo and Neodur showed very similar reactions and a moderate and a good level of resistance to yellow rust in terms of disease severity and infection type respectively. The recombinant inbred lines were characterized by a wide range of IT and DS values with statistically significant differences, and this finding indicates that resistant loci could be carried by both parents of this segregating population.

N. QTLs	Chrom.	Year	LOD	R <sup>2</sup>
1	2A	2019	3.1-5.4	0.11-0.18
2	2B	2018	3.9-8.3	0.13-0.26
1	4A	2018/2019	2.9-3.8	0.10-0.13
1	6B	2018/2019	2.4-3.6	0.08-0.12
1	7B	2018/2019	3.3-5.8	0.11-0.19

Six significant QTLs were identified considering the two years of evaluation. Interestingly, three out of them, on chromosomes 4A, 6B and 7B, were consistent in the two years. The QTL on chromosome 4A was contributed by Cirillo, while the ones on chromosomes 6B and 7B by Neodur. LOD values were between 2.4 and 8.3, with percentages of explained phenotypic variation ranging from 8% to 26%.

## Genome-wide association mapping in wild emmer wheat for pre-breeding activities

**Wild emmer wheat collection** (285 accessions). Principal Coordinate and Structure Analysis identified two well defined groups within the collection. Compared with geographical coordinates, the two recognized groups mostly correspond to the known wild emmer races, the North-Eastern (NE) one, represented by accessions originating from North-Eastern region of the Fertile Crescent (Turkey, Iraq, Iran), and the Western (W) one represented by accessions from the Western region of the Fertile Crescent (Israel, Jordan, Lebanon, Palestine, Syria). A number of identical accessions were identified, most sharing the same GPS origin coordinates.



PCoA analysis. Red dots correspond to accessions coming from Turkey/Iraq/Iran, while light blue dots represent accessions originating from Israel, Lebanon, Syria.

The collection was evaluated at **seedling stage for resistance to races PSTv-14, PSTv-37 and PSTv-40**. Tests were conducted in growth chamber at the University of Minnesota, using differential lines as controls.

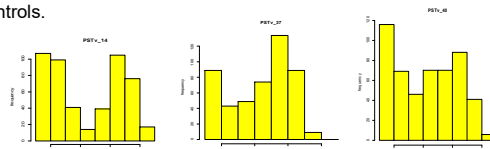


Figure 4. Distribution of scoring values for the three races

A **genome-wide association study (GWAS)** based on a Mixed Linear Model which considered Kinship relationships between accessions, and possibly heading date as covariate.

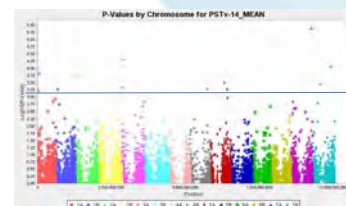


Figure 6. An example of Manhattan plot for one of the considered trait (Pstv14 resistance)

The collection was evaluated at **adult stage, in natural field conditions in Southern Italy for two years for Infection Type (IT) and Disease Severity (DS)**, with susceptible lines to increase the inoculation.

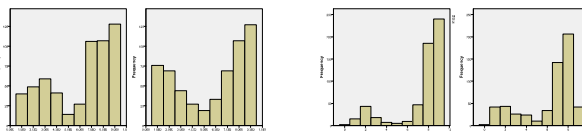


Figure 5. Distribution of scoring values for IT and DS in 2018 and 2019 field experiment

Scattered MTA plots were obtained as expected due to the fast LD decay.

The GWAS identified a number of resistance loci widespread into the genome, with **many specific loci for race specific resistance, and some conserved loci across the two years for open field resistance**.

## Conclusions

Through a survey on Zavitan genome annotation, for most of the loci we were able to identify candidate genes related to defense within 1Mb around the position of the MTA peak SNPs. Mostly they belong to these categories: resistant like genes, genes for membrane components encoding protein glycosylases, serine/threonine kinases, aminoacid transporters, genes for components of the intracellular signal transduction pathway...

Due to the continuous plant-pathogen co-evolution, the identification of new sources of resistance is needed. Only the exploration of large and diverse tetraploid wheat collections can ensure the identification of resistant loci to feed pre-breeding programs. On the other hands, finding resistance alleles in elite gene pool allows the fast availability of resistant cultivars for cultivation, and an easy transfer of the resistance to other cultivars of interest. The wild emmer wheat collection and the durum wheat RIL population led to the identification of non-overlapping QTLs, confirming the need to explore very diverse genetic resources.