

Melon Germplasm from Tunisia with Immense Breeding Value

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Melon (*Cucumis melo* L., $2n = 24$) is a morphologically diverse horticultural crop of high nutritional value and economic importance in Tunisia and the Mediterranean regions. The geographical position of Tunisia and range of climatic conditions, from humid in the north to arid in the south, have contributed to melon diversity in terms of landraces and wild genetic resources. Melons are cultivated in various areas in the country and largely consumed in summer, as a fresh fruit, appreciated for its sweetness.

Melon classification is a topic of great interest. It has been historically organized from a botanical viewpoint with the recent designation of three subspecies, *ssp. agrestis*, *ssp. melo*, and *ssp. meloides* (Endl et al., 2018). Botanical groups (*varietas*) have been defined within *ssp. agrestis* and *ssp. melo* (Pitrat et al., 2000). Melon may also be organized in a horticultural framework according to the International Code of Nomenclature for Cultivated Plants (Brickell et al., 2009). Burger et al. (2010) defined 16 Horticultural Groups within the two subspecies *agrestis* and *melo*. Pitrat (2016) later described 19 Horticultural Groups without reference to the subspecies. Horticultural Groups *Cantalupensis*, *Flexuosus*, *Inodorus* and *Reticulatus*, as defined by Burger et al. (2010), are the most important in Tunisia (Chikh-Rouhou et al., 2021c).

The agronomic traits and fruit characteristics of Tunisian landraces are similar to the commercial types demanded by Mediterranean markets, which facilitates breeding programs directed at this market. They also carry adaptation to the range of environments and cultivation methods in Tunisia, which suggests this diverse germplasm could be a great resource for melon breeding in a changing environment. In this paper, the local genetic resources of melon are reported and the breeding programs in which they are involved are

summarized to demonstrate the potential of Tunisian melon landraces as a valuable genetic reservoir and the need to plan strategies for its conservation and utilization in breeding programs.

Melon collections (Figure 1) at the Regional Research Centre on Horticulture and Organic Agriculture (CRRHAB) Tunisia were initiated in 2014. Accessions were collected and characterized for morphological traits (Chikh-Rouhou et al., 2021c; Trimech et al., 2013) and molecular markers (Trimech et al., 2015). Genetic diversity and population structure of the Tunisian melon collection were characterized by combining phenotypic and molecular data; specific alleles related to agronomic traits of interest were detected in two landraces, which constitute a potentially valuable gene pool for melon breeding (Chikh-Rouhou et al., 2021b).

Tunisian melon landraces have been identified as highly resistant to many biotic stresses such as powdery mildew (Chikh-Rouhou et al., 2020), *Fusarium* wilt incited by *Fusarium oxysporum* f.sp. *melonis* (FOM) (Chikh-Rouhou et al., 2018, 2021a), and melon aphid (*Aphis gossypii*) (Chikh-Rouhou et al., 2019). These studies combined phenotypic and molecular strategies to identify resistant accessions (Table 1). Molecular markers tightly linked to FOM and melon aphid resistance genes, *Fom-1* and *Fom-2*, and *Vat*, respectively (Oumouloud et al., 2012, 2015; Dogimont et al., 2009), and controlled inoculations were used to determine melon landraces resistant to FOM and melon aphid (Chikh-Rouhou et al., 2021a, 2019, 2018). Thirteen of the 27 landraces carried *Fom-1*, confirming their resistance to FOM races 0 and 2; two of them were also resistant to FOM race 1 (Table 1). Two accessions of *Inodorus* and *Flexuosus* groups with a high level of resistance to the most virulent race 1.2 of FOM

have been reported; the number of accessions resistant to race 1.2 is very low, and almost all of them belong to Groups Makuwa and Conomon (Chikh-Rouhou et al., 2010, 2011), which are common to parts of India and East Asia. Thus, the two FOM 1.2-resistant Tunisian landraces are very promising resistance sources to stem losses from this race, but further characterization of resistance to FOM 1.2 in these two landraces is needed before it can be incorporated into melon breeding programs. Resistance to race 1.2 is complex, controlled by multiple recessive genes with epistatic effects, which make selection difficult (Chikh-Rouhou et al., 2011; Perchepped and Pitrat, 2004).

Regarding resistance to melon aphid, Chikh-Rouhou et al. (2019) reported one of 15 landraces evaluated with the *Vat* gene, which confers resistance to melon aphid colonization and the viruses transmitted by this aphid (Table 1). This accession is also promising because several genomics studies focused on the region containing *Vat* showed that the density of host plant resistance genes in melon is highest in this region (Garcia-Mas et al., 2012). Thus, 28 genes of the NLR family have been identified in a 1-Mb region containing *Vat* (González et al. 2014), as well as the resistance genes to powdery mildew incited by *Podosphaera xanthii* (Yuste-Lisbona et al., 2001) and cucumber vein yellowing virus (CVYV) (Ibn Oaf, 2012), the *Fn* gene (Pitrat and Lecoq, 1984), which triggers plant necrosis in response to some isolates of zucchini yellow mosaic virus (ZYMV), and the quantitative trait loci (QTL) *FomV-2*, which confer partial resistance to FOM race 1,2 (Perchepped et al., 2005).

Particular attention should be given to landrace Chamem, an Ananas type, that carries *Vat* and *Fom-1* (Chikh-Rouhou et al., 2019; Chikh-Rouhou et al., 2021a) and was also found resistant to *P. xanthii* race 2 (Chikh-Rouhou et al., in preparation). It is a potential landrace with immense value as a donor of multiple pest resistances for melon breeding programs to develop commercial melons of Ananas type, which are highly appreciated not only in Tunisia but in other Mediterranean countries. Most reported sources of resistance to *P. xanthii* come from India (e.g., PI 124112, PI 414723, PI 134198, PI 313970) and a few have been reported in Groups Momordica and Acidulus (Nunés et al., 2017), but their agronomic and fruit characteristics are usually unsuitable for the Mediterranean market, which makes them problematic as sources of powdery mildew resistance.

Comparative studies of disease resistant and tolerant melon genotypes for differences in their microbiomes are ongoing at CRRHAB in order to identify key microorganisms potentially involved in modulating the defensive/resistance responses that may be taken into account in future breeding

programs. A recent study of Aydi-Ben-Abdallah et al. (2021) on fungal and bacterial rhizosphere microbiomes associated with selected Tunisian melon landraces demonstrated the following: 1) genotypic differences for quantum and diversity of their microbiomes and 2) soil microbial structure–melon genotype interactions that may be exploitable for development of melon lines with high-level, stable yield potential by inclusion of holobiont selection in breeding programs. All these investigations have led to the identification of local (Tunisian) germplasm of high interest for their resistance to biotic stress, fruit quality, and agronomic behavior. Hybrids from the promising parents identified could improve heterosis for quality traits and yield in melon. Breeding of local hybrid varieties was initiated in 2019 at CRRHAB and newly created F₁ hybrids resistant to Fusarium wilt are currently under evaluation in different sites in order to select the best ones for quality and yield. These lines are also the subject of diallel, heritability, GCA (General Combining Ability), and SCA (Specific Combining Ability) analyses in order to optimize selection of elite materials for Tunisia.

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Table 1. Horticultural characteristics–Horticultural Group, total soluble solids (°Brix), external aroma–of 27 Tunisian melon landraces and their resistances to *Fusarium* wilt incited by *Fusarium oxysporum* f.sp. *melonis* and melon aphid (*Aphis gossypii*), as determined by molecular markers.

Accession		Resistance marker				
Code	Name	Group ^z	°Brix	Aroma ^z	Fusarium wilt ^x	Melon aphid
TUN-1	Maazoun Chott-Mariem	Inodorus	9.8 ± 0.6	no	<i>Fom-1/fom-1</i>	<i>vat</i>
TUN-2	Maazoun Menzel Chaker	Inodorus	10.2 ± 1.1	no	<i>Fom-1/Fom-1</i>	<i>vat</i>
TUN-3	Maazoun Mehdi (MM2009)	Inodorus	11.0 ± 1.9	no	<i>Fom-1/Fom-1</i>	<i>vat</i>
TUN-4	Maazoun Fethi	Inodorus	10.9 ± 0.5	no	S ^y	<i>vat</i>
TUN-5	Fakous (FL)	Flexuosus	4.7 ± 0.5	no	<i>Fom-1/fom-1 Fom-2/fom-2</i>	–
TUN-6	Fakous Salem 2014	Flexuosus	–	–	<i>Fom-1/fom-1</i>	–
TUN-7	Trabelsi	Inodorus	9.7 ± 0.3	no	S	<i>vat</i>
TUN-8	Galaoui	Reticulatus	10.1 ± 0.4	yes/no	S	<i>vat</i>
TUN-9	Dziri (DZ P5 2011)	Inodorus	9.6 ± 0.9	no	<i>Fom-1/Fom-1</i>	<i>vat</i>
TUN-10	Lobneni	Reticulatus	9.5 ± 0.2	yes/no	<i>Fom-1/Fom-1</i>	–
TUN-11	Arbi	Inodorus	–	–	S	–
TUN-12	Horchay	Chate	7.4 ± 0.4	no	<i>Fom-1/fom-1 Fom-2/fom-2</i>	–
TUN-13	Arbi 1	Inodorus	9.8 ± 0.1	no	S	<i>vat</i>
TUN-14	Arbi 2	Inodorus	8.4 ± 0.2	no	<i>Fom-1/Fom-1</i>	–
TUN-15	Arbi 3	Inodorus	10.2 ± 1.1	no	S	–
TUN-16	Sarachika	Inodorus	9.8 ± 0.5	yes/no	<i>Fom-1/fom-1</i>	<i>vat</i>
TUN-17	RD	Cantalupensis	11.8 ± 1.1	yes	S	<i>vat</i>
TUN-18	Rupa	Cantalupensis	9.7 ± 0.7	yes	<i>Fom-1/fom-1</i>	<i>vat</i>
TUN-19	Chamem (Ananas type)	Reticulatus	10.0 ± 0.2	yes	<i>Fom-1/Fom-1</i>	Vat
TUN-20	HTM Kairouan	Reticulatus	–	–	S	–
TUN-21	Acc Jendouba	Inodorus	–	–	S	–
TUN-22	Dziri (Menzel Kamel)	Inodorus	–	–	S	–
TUN-23	Ecotype arbi Dz	Inodorus	–	–	S	–
TUN-24	Maazoun (Kairouan)	Inodorus	9.5 ± 0.6	no	S	<i>vat</i>
TUN-25	Asli	Inodorus	11.43 ± 0.5	no	S	<i>vat</i>
TUN-26	Stambouli	Inodorus	8.93 ± 1.1	no	<i>Fom-1/Fom-1</i>	<i>vat</i>
TUN-27	V4 autoféc	Inodorus	11.5 ± 1.3	no	S	–

^zBurger et al. 2010

^xChikh-Rouhou et al. 2021a

^yS: susceptible



Figure 1. Diversity of Tunisian melon landraces. (Photo H. Chikh-Rouhou)