

Preliminary results on fingerprinting of lemon genotypes tolerant to mal secco disease by RAPD markers

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Abstract: We investigated the possible utility of randomly amplified polymorphic DNA (RAPD) markers in our lemon (*Citrus limon* Burm. F.) breeding program emphasizing the excellent quality and tolerance to mal secco disease caused by *Phoma tracheiphila* Kanc. et Ghik. The genotypes studied included 12 advanced selections that were previously shown to be resistant or tolerant to this disease and 4 lemon varieties (“Küt diken 79”, “Tuzcu 05-Yediveren Küt diken”, “Finike Yerli Yuvarlak”, “Antalya Yerli Yuvarlak”). DNA characterized the 16 lemon genotypes. We scored 350 bands generated by 48 selected RAPD primers. 38.85% of these were constant corresponding to monomorphic loci while 61.14% of the bands were polymorphic. The number of bands for each primer varied from 2 to 15 with an average of 7.3 bands per primer, the sizes of the amplified DNA bands ranged from 250 to 3000 bp. Primers differed in their capacity to detect polymorphism and total proportion of polymorphic products ranged from 12.5% to 100%. The analysis of 16 lemon genotypes using 48 RAPD primers allowed us to distinguish all genotypes except “Tuzcu 896” and “Tuzcu 897” which were selection of “Tuzcu 9N Aklimon”. The dendrogram indicated that the genotypes can be separated into two major groups with a similarity value of 0.79. These results show that utilization of RAPDs can offer great benefits to our lemon breeding program in several ways, such as identifying genotypes and studying the genetic similarity among genotypes. The future work can be extended to develop RAPD markers linked to gene(s) conferring tolerance to mal secco.

Key words: DNA fingerprinting, cluster analysis, *Citrus limon*, lemon, mal secco, RAPD markers.

Abbreviations: ISSR, inter-simple sequence repeats; RAPD, randomly amplified polymorphic DNA; RFLP, restriction fragment length polymorphism.

Introduction

Lemon (*Citrus limon* Burm. F.) belongs to the most important Citrus in the Mediterranean area. Mal secco disease, caused by *Phoma tracheiphila* Kanc et Ghik, is one of the most important problems for lemon growers in this region. Although the disease occurs in Italy, Spain, France, Greece, Turkey, Tunisia, Algeria, Cyprus and the Eastern Black Sea coast area of Russia, it is most prevalent in the Eastern-Mediterranean region where it causes heavy enough losses to threaten lemon culture.

In Turkey, mal secco was first seen in small areas in 1933. After the 1940s, however, the disease spread rapidly with the expansion of lemon cultivation (TUZCU et al., 1989). As is the case in other Mediterranean countries, the development of improved lemon cultivars with tolerance or resistance to mal secco is an important breeding objective in Turkey. To achieve this goal, a rig-

orous breeding program has been developed at the Faculty of Agriculture of the Çukurova University (Adana, Turkey), employing scion selection, hybridization and mutation breeding. Several mal secco tolerant genotypes have been identified, for example “Antalya Yerli Yuvarlak” and “Finike Yerli Yuvarlak” (TUZCU et al., 1989). However, the quality of these tolerant genotypes is not as desirable as that of “Feminello” (i.e. “Küt diken”). Thus, the breeding program has focused on combining tolerance to mal secco with excellent quality. Currently, there are twelve advanced selections from this breeding program. These genotypes have passed the juvenile stage and can provide data on mal secco tolerance as well as several other horticulturally important traits (TUZCU et al., 1992).

During the past few years, fruit breeders have employed molecular markers (LUBY & SHAW, 2002) for mapping, fingerprinting cultivars, marker-assisted selection, and other uses. Such analyses shift focus from

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