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**THE GENETIC DIVERSITY OF APULIAN APRICOT GENOTYPES  
(*Prunus armeniaca* L.) ASSESSED USING AFLP MARKERS**

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**Abstract:** Apricot is an important crop in Italy and, especially in Southern regions, in the last five years numerous plantings using new cultivars and appropriate cultural management have been established. The cultivars available were created in different environments (USA, France, New Zealand, etc), they then often show low adaptability to Italian conditions. However, in the South of Italy, it is still possible to safeguard and to exploit a considerable amount of the apricot genetic variation available in ecotypes often characterised both by useful bio-agronomic traits and by good environmental adaptation. These genetic materials could be used in breeding programs aimed at broadening the harvest period and obtaining high fruit quality and resistance to the main biotic and abiotic stresses.

**Key Words:** Apricot, Genetic Diversity, Molecular Markers, Ecotypes

**INTRODUCTION**

*Prunus armeniaca* L. is one of the most important *Prunus* species grown in the world. More than 80% of the world's apricot production is restricted to the Mediterranean regions. The cultivars used in Italian plantations often had their origins in other environments, and show low adaptability to Italian conditions [1].

The Mediterranean region, more specifically the Apulian region, is a secondary centre of apricot species diversification. It provided an opportunity for breeders

to carry out a project on the collection, conservation, evaluation and sustainable use of apricot germplasm. The germplasm originally consisted of numerous ecotypes belonging to wider populations conserved for their capacity to adapt to local environments [2]. The study of genetic diversity in apricot, as well as in other fruit species, is important not only for evolutionary studies but also from the point of view of plant breeding and germplasm conservation. The relationships between genotypes are evaluated through the estimation of genetic distances or coefficients of similarity based on morphological, biochemical or molecular markers.

Molecular markers are currently being applied in genetic diversity studies because, unlike morphological characteristics, they are not affected by environmental variation. Restriction fragment length polymorphism (RFLP) has been used for various species including apricot [3, 4], but this technique is laborious and involves expensive and radioactive/toxic reagents. Other molecular markers, such as microsatellites and random amplified polymorphic DNA (RAPD) [5] have been developed for *Prunus* species to prepare a genetic map or to analyse the genetic diversity of *Prunus* rootstocks.

Amplified fragment length polymorphism (AFLP) permits the generation of hundreds of informative genetic markers [6]. AFLP markers have been used to detect genetic variation among and within populations [7, 8] and determine the genetic structure and differentiation of populations [9, 10].

This paper shows the results obtained from samples collected from several sites of the Apulian region with the aim to collect or maintain *in situ* genetic resources of apricot. Thirty-nine genotypes of apricot (5 cultivars and 34 ecotypes) were identified and studied for different bio-agronomical traits. The genotypes were also differentiated as regards their molecular polymorphisms revealed by means of AFLP analysis.

## **MATERIALS AND METHODS**

### **Plant material**

Plant material consisted of five cultivars and 34 ecotypes, mainly represented by isolated individuals, ranging from 20 to 80 years of age. The identification of genotypes was done in different areas of the Apulian region. These genotypes have been characterised over a two years period recording their main bio-agronomic traits: flowering time, ripening time, and fruit size.

### **DNA extraction and AFLP analysis**

Total DNA was extracted using fresh leaf tissues (5 g) according to Sharp [11] with minor modifications. The AFLP procedure was performed following the protocol described in Vos [12]. The AFLP amplification reaction was performed according to the AFLP small genome plant protocol (AB Corp., Norwalk, Connecticut, USA). Capillary electrophoresis was carried out on an ABI PRISM 3100 (AB Corp., Norwalk, Connecticut, USA).

### Data analysis

The raw data were processed using “Genescan” software. All scored bands were analysed using “Genotyper” software. A matrix of raw data was constructed on the presence (1) or absence (0) of each polymorphic DNA fragment in all the genotypes tested. A phenetic tree was obtained by means of the NTSYS software [13]. Genetic similarity was measured using the test of Nei and Li [14].

## RESULTS AND DISCUSSION

This study permitted the identification 34 apricot ecotypes in the Apulian region (South Italy).

Bio-agronomic data collected over two years (2000 and 2001) showed a high degree of variation between genotypes. Flowering time ranged from February to April and ripening time from May to September. Moreover, the genotypes under consideration produced of different fruit size (small, medium and large) commonly showing a high degree of self-compatibility. The variability detected among genotypes in relation to bio-agronomic traits was also confirmed by molecular analysis. By means of molecular markers, high variability has been observed in *Prunus* by other authors [4, 5, 15-19].

In this paper, the AFLP analysis using 4 primer combinations (see Table 1) produced 409 amplified fragments. Only 267 bands, ranging from 35 to 500 bp, were polymorphic (61%).

Tab. 1. Polymorphism rates related to the four primer combinations

Primer combination	Number of bands	Polymorphic bands	Polymorphism (%)
E-ACA/M-CAT	153	119	78
E-ACA/M-CAG	78	52	67
E-ACA/M-CAA	140	80	57
E-ACT/M-CAA	38	16	42
	Total 409	Total 267	Total 61

As shown in the dendrogram, the coefficient of similarities based on AFLP fragments revealed genetic diversity between the apricot genotypes under consideration (Fig. 1). In the dendrogram, it is possible to distinguish five groups. Only two genotypes aggregated into a cluster at a higher level of similarity (0.98) (Dspv14 and Dspv22); all the other genotypes aggregated into the five clusters at lower similarity values, ranging from 0.50 to 0.80.

Genotypes from different areas of the Apulian region are included in each of these clusters. Thus, no correlation seems to exist between geographical between

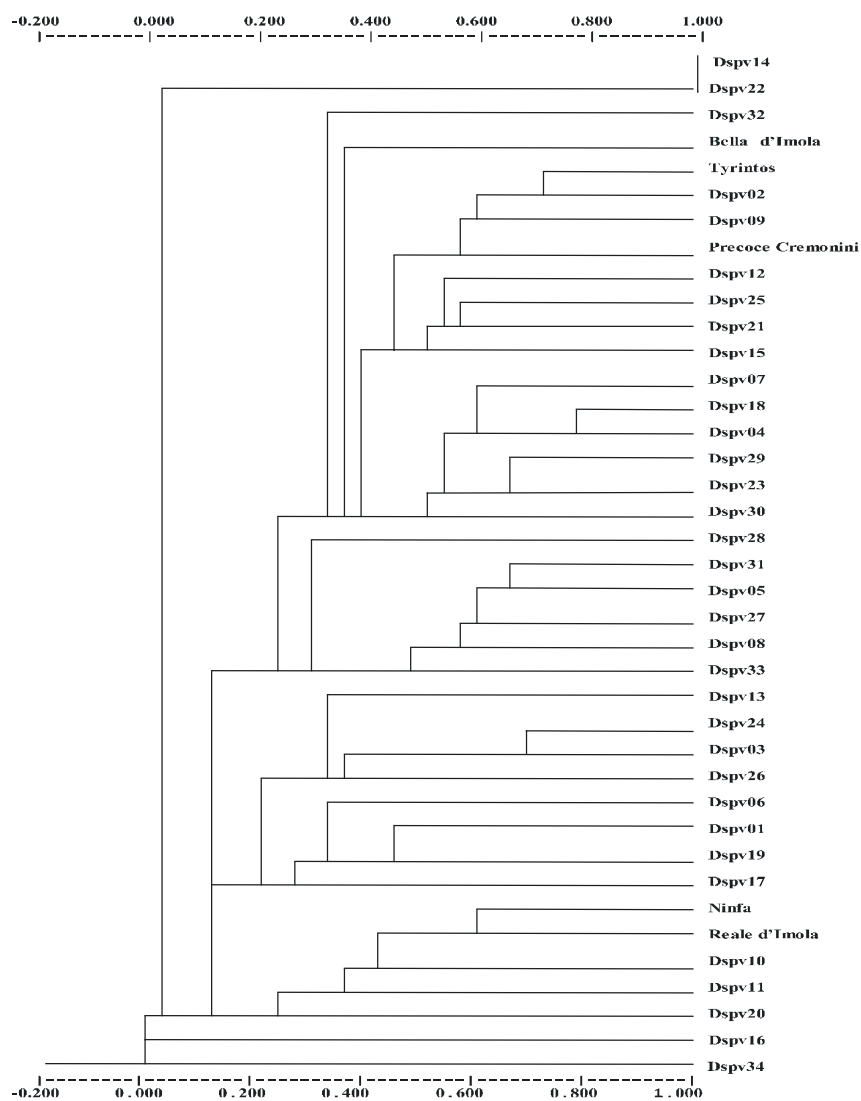


Fig. 1 Dendrogram of apricot genotypes based on AFPL data using Nei's genetic distance matrix of similarity and the UPGMA clustering method.

distribution and bio-agronomic data, in accordance with the molecular analysis. In addition, genotypes with different flowering and ripening times, as well as fruit size, are included in each of these clusters, indicating no relationship between cluster formation and bio-agronomic characteristics. The five cultivars (from different regions) which have been included in this analysis appear to be aggregated separately in different clusters; it might suggest some relationship

these cultivars and some ecotype grown in Apulia. The general pattern of aggregation into clusters indicates large variability among the considered genotypes.

The characterised germplasm represents an important source of variation for the Apulian environment, and it might be useful for breeding programs aimed at the selection of new self-compatible cultivars with good bio-agronomic performances.

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