AFLP GENETIC CHARACTERIZATION IN ASPARAGUS OFFICINALIS DOUBLED-HAPLOID (DH) CLONES COLLECTION

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Cultivated asparagus (Asparagus officinalis L., Liliaceae 2n=20) is a perennial dioecious species, for its tasty stems it is an edible crop of high economic value. Sex is determined by gene(s) on a pair of homomorphic sex chromosomes with homogametic (XX) females and heterogametic (XY) males. However, rarely andromonoecious individuals are observed among the males. These are of XY constitution but able to self-fertilize and produce fertile berries. The F₁ progeny of these selfings segregate in the ratios 1XX: 2XY:1YY, producing the so-called supermales (YY). All-male populations of asparagus crop are widely considered superior to mixed populations, due to higher yield, no contamination of the field by germinating berries and longevity. While they are generally weak and of low productivity themselves, YY-plants are highly valued for the production of hybrid seeds, as all the progeny from crosses with female XX-plants will be of the male XY constitution.

An alternative way to produce supermales is the production of doubled-haploids (DH) via anther culture. The regenerated plants should be of haploid constitution but usually double their chromosome number spontaneously, or induced by chemicals, at a very early stage, yielding doubled-haploid XX and YY plants. The in vitro anther culture technique has been suggested as the best way to obtain doubled haploid female and male clones of asparagus which can be used as parents of F₁ all-male hybrids. This technique has been applied in Italy during the last twenty-five years and has allowed to release commercially F₁ all-male hybrids.

The present report focused on the genetic characterization of several double haploid accessions, part of an important collection conserved at the C.R.A. Research Institute for Vegetable Crops, section of Montanaso Lombardo. Genetic diversity was analyzed using AFLP molecular markers strategy; forward primers of the nine primer-pairs used were labelled with ABI PRISM fluorescent dye 6-FAM, and the PCR reactions were run on ABI PRISM. The automated sequence system ABI PRISM combined with fluorescent labelling of expected fragments has been applied as an alternative to radioactivity detection using [³²P] or [³³P]-labelling. This technology provides an automatic and rapid sizing of the fragments through the use of specific internal size standard (GS500-Liz) and allows analysis of fragments. The data collected from each sample were automatically analysed by GeneMapper Analysis Software.
The genetic diversity were estimated; a matrix of presence/absence of DNA fragments has been used for the comparison of the accessions in order to obtain the coefficients of genetic similarity (Dice, 1945). The coefficients have been utilized for the UPGMA (Unweighted Pair Group Method Averages) analysis useful for obtaining a dendrogram among accessions (PAST ver. 1.12 software).

The genetic relationships among double haploid lines together with the morpho-agronomic data should be now utilized in the choice of parental genotypes for F₁ hybrid constitution.