*Proceedings of the 53rd Italian Society of Agricultural Genetics Annual Congress*
*Torino, Italy – 16/19 September, 2009*
ISBN **978-88-900622-9-2**

**Poster Abstract – 3.11**

# CAFFEOYLQUINIC ACID BIOSYNTHESIS STRUCTURAL GENES IN *CYNARA CARDUNCULUS* L.: ISOLATION, HETEROLOGOUS EXPRESSION AND GENOME MAPPING

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*secondary metabolites, caffeoylquinic acid biosynthesis, SNP marker development, genetic mapping*

Plant secondary metabolites are highly evolved compounds performing different functions, and have been widely exploited from food to medicine. A constant supply of phenols, a class of secondary metabolites, provides preventive and defensive mechanisms to reduce the risk of chronic diseases in human beings; among them caffeoylquinic acids (CQAs) have attracted a growing academic and industrial interest in recent years. In various pharmacological test systems, extracts of plants with a high content of CQAs exhibit hepatoprotective, anticarcinogenic, antioxidative, antibacterial, anti-HIV activities as well as the ability to inhibit cholesterol biosynthesis and LDL oxidation. However, at present, fragmentary information are available on CQAs biosynthesis in plants.

*Cynara cardunculus* L. includes globe artichoke (var. *scolymus* L.), cultivated cardoon (var. *altilis* DC.) and their progenitor wild cardoon [var. *sylvestris* (Lamk) Fiori] and represents a very interesting model species for studying CQAs, due to the exceptionally high natural content and diversity of these compounds. In previous works, the full-length cDNAs of two transferases (HCT and HQT) and a hydroxylase (C3'H) have been identified and characterized by our group and they have been positioned on genetic maps.

Here we report on the analysis of a set of 19000 globe artichoke unigenes (made available by the Compositae Genome Project, http://compgenomics.ucdavis.edu/), that allowed the identification of 7 genes (i.e. a 4-coumarate:coenzyme A ligase, a cinnamate-4-hydroxylase, a MYB12 like gene and four transferases) putatively involved in CQA biosynthesis. The allelic forms of these genes were sequenced in the parental genotypes (the globe artichoke clone 'Romanesco C3' and a genotype of cultivated cardoon 'Altilis 41') of a $F_1$ progeny used for the construction of a linkage map based on a two-way pseudo-testcross strategy. SNPs were detected and the analysis of their segregation made possible gene positioning in the genetic map. Their linkage group distribution will be the starting point for gaining a better knowledge of the genetic bases (i.e. Quantitative Trait Loci, QTL) of the CQA biosynthesis.

The full-length cDNAs of the four identified transferases, characterized by a high similarity with the *C. cardunculus* acyltransferases (HCT and the HQT), have been isolated by means of 5' and 3' RACE technique and heterologously expressed in *E. coli.* HPLC analyses of the reactions catalysed by these isolated enzymes are at present in progress, in order to investigate their role in CQA biosynthesis.