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# **A Multidisciplinary Approach to Preserve and Value the Chestnut Heritage**

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## **Abstract**

**In Italy the spread of chestnut (*Castanea sativa* Mill.) has favored the evolution of a rich varietal heritage in different pedoclimatic areas. Conservation of this wide germplasm is very important to save valuable genotypes, because they may retain special adaptative and technological traits and so meet the demands of the market that nowadays requires, more and more, typical products of high quality. The preservation and valorization of this heritage can be achieved through a multidisciplinary approach. Genetic, morphologic, chemical and sensory analyses allow to define the properties and the characteristics of each chestnut cultivar and can be helpful in choosing the most proper uses of the nuts. In this work, we describe our experience on chestnut characterization aimed to select plant material to be exploited in the different growing areas and show how a multidisciplinary approach greatly improves the efficacy of the characterization and the reliability of the evaluation of the germplasm. The final aim is to provide growers with selected plant material able to yield nuts suitable for the fresh market or for processing and offer to consumers high quality products from vocated areas.**

## **INTRODUCTION**

The preservation of the chestnut germplasm from the genetic erosion due to the changes in the socio-economic structure of rural areas and specific pathogen attacks (Arnaud *et al.*, 1997; Sartor *et al.*, 2009) is an important objective in the agro-biodiversity conservation strategy (CBD 2002). From a socio-economic point of view, chestnut can play an important role in promoting local identity and social cohesion as well as in helping to preserve the landscape; where the cultivation of this species is well established. The rediscovery of traditional local genotypes has the potential to form the basis of initiatives that can be developed for the benefit of the local communities.

The evaluation and selection of the cultivars, on the basis of quality traits, is essential to give back to the local chestnut growing areas, the uniqueness and specificity that have always characterized the mountainous lands; moreover, traceability is nowadays a requirement to protect the chestnut European productions and their superior quality.

A better knowledge of the chestnut germplasm is the starting point in order to reorder the genetic resources, to preserve biodiversity, environment and historical memory of a place, to promote and value local products. This goal can be achieved only through a multidisciplinary approach. Morphological and genetic analyses are necessary to correctly identify and describe cultivars and genotypes, in order to provide useful and reliable information for the identification of plant material in orchards and nurseries (Mellano *et al.*, 2012, Torello Marinoni *et al.*, 2013). Chemical and sensory analyses are necessary to assess quality standards for different kinds of chestnuts, destined by tradition to specific culinary uses (Mellano *et al.*, 2009).

The purpose of this work was to give an overview of our experience on chestnut characterization applied in Northern Italy. We briefly reported some case studies in Piedmont Region: Capanne di Marcarolo Natural Park, Val Grande National Park, Valli di Lanzo Mountain Community, Val Pellice Mountain Community.

The Capanne di Marcarolo Natural Park is a protected environment, located in a mountainous area of the Appennino Ligure-Piemontese, which still preserves features of wilderness

and wildlife. Val Grande National Park, a few steps from Maggiore Lake, East Piedmont, is the largest wilderness area in Italy, with old chestnut trees and a most valuable biodiversity. Valli di Lanzo Mountain Community joins many municipalities in the North-western part of Piedmont and its main purpose is to favor the development of the valleys and the preservation of their cultural and environmental heritage; in this area the chestnut was present with a rich varietal heritage, though still undervalued because not adequately characterized and selected. Val Pellice. Mountain Community joins municipalities in the South-western part of Piedmont, and it is rich of excellent chestnut cultivars, such as the well known 'Marrone'.

## **MATERIALS AND METHODS**

The steps for the rescue and valorization of the germplasm can be summarized in the following way:

1) Survey and plant positioning: plants are selected in collaboration with local technicians and farmers, labelled with a code and positioned on a map by GPS (Global position system);

2) Morpho-functional description of the selected plants: phenological (bud break, times of male and female flowering, harvesting date), morphological (height of the tree, trunk diameter) traits and health conditions are recorded and included in a data sheet;

3) Sampling for laboratory analyses: leaves, flowers, and nuts are collected for morphological characterization, leaves and buds for DNA analysis, nuts for carpology and sensory evaluation;

4) Genetic analyses: microsatellites or simple sequence repeats (SSR) are the most used molecular markers for the identification and characterization of chestnut germplasm, allowing a reliable cultivar identification;

5) Morphologic and carpological analyses: the principal descriptors to be used are included in the descriptor list for chestnut of the International Union for the Protection of New Varieties of Plants (UPOV 1989) and of the Inventory of Chestnut Research Germplasm and References (Bounous *et al.*, 2002). Based on our experience, the most relevant carpological traits are: size (number of nuts per Kg), nut shape, nut colour, nut hairiness, percentage of polyembryony, pellicle intrusion, pellicle adhesion to kernel; meanwhile the leaf and flower traits more useful are: petiole length, length/width ratio of foliar blade, male flower type and length of unisexual catkins;

6) Chemical analyses: in order to evaluate nut quality, seeds are analyzed for starch and total dietary fiber (TDF) (by enzymatic methods), for lipid content (by Soxhlet), for total soluble sugars (TSS) (by gaschromatography), for proteins (by Nitrogen Kjeldahl), for ashes (in oven at 550°C for 5h).

7) Sensory analyses: these analyses can be performed on chestnuts (fresh, boiled, roasted) or flour obtained from dried nuts. In general, nuts are evaluated by a group of 10-12 panelists using a specific sensory data sheet for the quantitative-descriptive test. The most important descriptors recorded are: ease of peeling, regularity of shape, colour, odour, sweetness, salty taste, bitterness, firmness, texture, aroma intensity. Descriptors are evaluated using a continuous, partially structured, scale (0-10). Panelists are also requested to give an edonistic judgment on flavour and overall quality.

8) Statistical analyses on the data set collected.

## **RESULTS AND DISCUSSION**

The research founded by the Natural Park Capanne di Marcarolo had the specific aim to describe and safeguard biodiversity. Thirty-eight old and grafted chestnut trees were collected and the DNA typing separated 22 genotypes. No accessions retrieved in other chestnut areas and analyzed in recent years by the Department were the same, for the markers examined. Morphological and chemical analyses allowed to describe in detail the chestnut production and showed that most of the cultivars were suitable for drying or for the production of flour. Moreover, in 2004 a collection of the chestnut genetic variability collected was established in the area of the

Park and a coppice was converted into an orchard, grafting the plants with local germplasm (Beccaro *et al.*, 2009)

The research founded by the National Park Val Grande had the specific aim to recover the landscape and biodiversity. For genetic analyses 33 plants were analyzed at 10 SSR loci, detecting 18 genotypes. DNA typing showed that the cultivars found in the Park are different from those present in the main cultivation areas in Piedmont. Morphological analyses allowed to select accessions with valuable quali-quantitative traits (Beccaro *et al.*, 2009)

The research founded by the Mountain Community Valli di Lanzo had the specific aim to select clones for the fresh market. First of all we proceeded with genetic analyses, in order to define the genetic profiles of the main cultivars spread across the territory. Therefore 57 plants were located and analyzed at 11 SSR loci: Twenty-seven different genotypes were found, allowing to identify cases of synonymy and homonymy and new genotypes (26) present exclusively in the investigated territory. A large variation above all in size, pellicle intrusion and ease of peeling was highlighted, with morphological analyses. Some genotypes of interest were selected: one for nut size, one for absence of pellicle intrusion, two for ease of peeling. By means of sensory analyses the 3 cultivars considered best by the consumers were identified. An important result of this work was the registration of 2 local cultivars ('Bojana' and 'Viri') in the Italian National Catalogue.

The research carried out in Val Pellice and funded by EU (MANCHEST project) had the specific aim of selecting clones for different uses. For genetic analyses, SSR markers provided a large amount of information on the genetic structure of this local germplasm; 17 plants were analyzed at 10 SSR loci and 10 genotypes were found (Torello Marinoni *et al.*, 2009). Chemical analyses showed a wide variability among cultivars as concerns nut composition. Starch and sucrose were the most abundant compounds, followed by total dietary fiber; proteins, lipids and ashes were present in lower amounts. Sensory analysis allowed the selection of a group of cultivars and clones particularly appreciated for their taste and aroma. Candying tests and sensory analyses provided information on the best clones of 'Marrone' to be propagated and used in new plantings.

## CONCLUSIONS

The aim of this paper is to show how a multidisciplinary approach can greatly improve the efficacy of the characterization and the evaluation of the local chestnut germplasm. According to the results, it is clear that the diversity present within the chestnut germplasm grown in Piedmont is very high and the differences are maintained also after processing. The chestnut cultivars of the Region can thus reach the market as a large array of commodities, representing a prized food for an increasingly large market sector.

The biodiversity described in the researches developed in North West Italy witnesses the history and the life of the people who lived in these areas and must be recovered, protected and improved. The selection of the best cultivars and clones for the different uses will favor the planting of orchards with improved plant material.

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