

Globe Artichoke and Cardoon

Sergio Lanteri¹ and Ezio Portis¹

¹ University of Torino, Di.Va.P.R.A. Plant Genetics and Breeding, sergio.lanteri@unito.it

1 Introduction

Globe artichoke (*Cynara cardunculus* var. *scolymus* L.) contributes significantly to the Mediterranean agricultural economy, with an annual production of about 750 Mt (more than 60% of global production) from over 80kha of cultivated land. Italy is the leading world producer (about 470 Mt), followed by Spain (188 Mt), France (52.5 Mt) and Greece (35 Mt). In southern Europe, artichoke production is an important component of regional economic stability and social development and, thanks to its long growth cycle, its cultivation provides employment almost the whole year round. Globe artichoke is also cultivated, although to a lesser extent, in the Near East (Turkey and Iran), North Africa (Egypt, Morocco, Algeria, Tunisia), South America (Argentina, Chile and Peru), and the United States (mainly in California), and its cultivation is spreading in China (55 Mt in 2005) (FAO data 2005: <http://faostat.fao.org/>). The major importing countries of the fresh product are France, followed by Italy and Canada. Spain is the leading exporter of processed product, being responsible for about 85% of the traded market (Bianco, 2007). The edible parts of the globe artichoke are the immature composite inflorescences (heads or buds, more formally referred to as capitula), and these are consumed worldwide as both a fresh and a canned delicacy (Figure 1). At flowering, leathery green bracts encase a purple-blue flower. Each plant produces small, medium and large capitula, with the largest formed at the apex of the central stem (primary head). The smaller capitula (secondary heads) develop on the lateral branches. Both spiny and non-spiny types are cultivated in different areas.

The cultivation of cardoon (*C. cardunculus* var. *altilis* DC) is much less widespread than that of the globe artichoke; it remains of regional importance in Spain, Italy and the south of France, where it is used in traditional dishes. The edible parts of the plant are the fleshy stems (Figure 2) which are typically collected in late

autumn-early winter. The leaves of the basal rosette are usually deeply divided, petiolate, very large (>50 x 35 cm), subcoriaceous, bright green and slightly tomentose on the upper surface, and white-tomentose on the lower one. Plant height can be over 2 m, and the flowers (blue, lilac or whitish) are grouped in globose capitula. Before cooking, the material is tied together, wrapped in straw, and/or buried for about three weeks in order to accentuate the flavour.

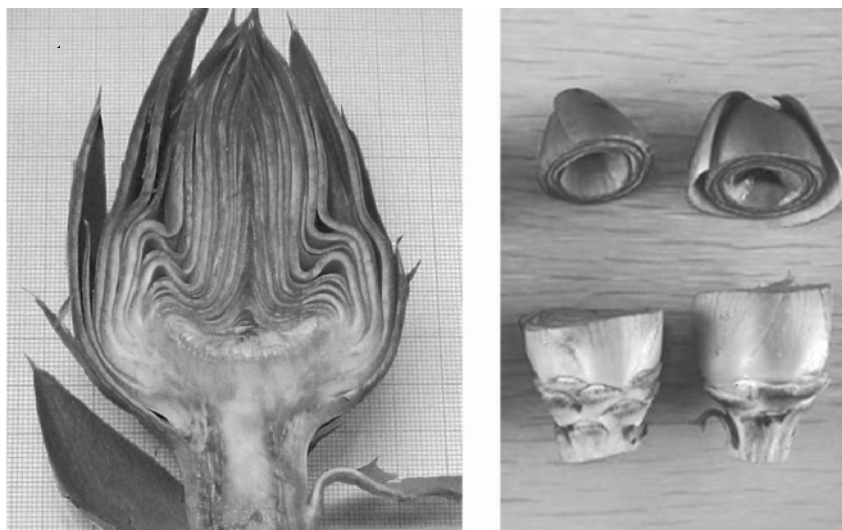


Fig. 1. Section of an immature capitulum of globe artichoke ‘Spinoso sardo’ (A) and the edible part of the heads (B).

The major (but not the only) use of globe artichoke and cardoon is for human food. Seed yield in cardoon is about 2 t/ha and up to 0.8 t/ha in globe artichoke (at 5% w/v moisture), about 25% of which is oil of good alimentary quality (Foti et al., 1999). The oil has (i) a high and well balanced content of oleic and linoleic acids, (ii) a low content of free acids, peroxides, saturated and linoleic acids, and (iii) a favourable α -tocopherol content, which provides a good level of protection against oxidation (Maccarone et al., 1999). Furthermore, the seed material left after the extraction of oil can be used as a component of animal feed. Cardoon has been identified as a candidate for the production of lignocellulosic biomass and paper pulp (Quilho et al., 2004; González et al., 2004). Both species have long been known to represent a significant source of biopharmaceuticals (Adzet and Puigmacia, 1985; Debenedetti et al., 1993; Slanina et al., 1993; Wagenbreth, 1996; Sevcikova et al., 2002; Wang et al., 2003), and recent trials have demonstrated that some of the end products can be produced in worthwhile quantities even when the crop is grown in environments unsuitable for normal commercial production (Matthes and Honermeier, 2007). The roots and rhizomes, used also for brew or infusion, provide a source of inulin, a demonstrated enhancer of the human intestinal flora; and the leaves provide a source of antioxidants such as luteolin and di-caffeoylquinic acid, which (i) protect proteins,

lipids and DNA from oxidative damage from free radicals (Gebhardt, 1997; Brown and Rice-Evans, 1998; Perez-Garcia et al., 2000), (ii) inhibit cholesterol biosynthesis and contribute to the prevention of arteriosclerosis and other vascular disorders (Kraft, 1997; Brown and Rice-Evans, 1998; Gebhardt, 1998; Pittlern and Ernst, 1998), (iii) inhibit HIV integrase, a key player in HIV replication and its insertion into host DNA (McDougall et al., 1998; Slanina et al., 2001), and (iv) have antibacterial activity (Martino et al., 1999). Extracts from the leaf and capitula can be used for the preparation of alcoholic drinks, beauty creams (Barbagallo et al., 2007), and milk coagulant used in the preparation of traditional cheeses such as the Algerian 'Djben' (Mouzali et al., 2004). It also is common as both an ornamental garden plant and source of cut flowers. The variously shaped capitula and the large blue or violet flowers are popular in floral compositions, and vase life ranges from 7 to 10 days depending on bud stage at harvest and the display conditions.



Fig. 2. Blanched stems of cardoon 'Gobbo di Nizza'.

2 Origin and Domestication

Cynara is a small genus belonging to the *Asteraceae* (daisy) family (Wiklund, 1992); it comprises seven species native to the Mediterranean basin - the crop complex *cardunculus*, which includes the globe artichoke, the cultivated cardoon and the wild cardoon (var. *sylvestris* (Lamk) Fiori), *C. syriaca* Boiss, *C. cornigera* Lindely (syn. *sibthorpiana* Boiss. et Heldr.), *C. algarbiensis* Cosson, *C. baetica* (Spreng.) Pau (syn. *alba* Boiss.), *C. humilis* L. and *C. cyrenaica* Maire & Weiller (Rottenberg and Zohary, 2005). The three *C. cardunculus* forms are fully cross-compatible with one another, and their F₁ hybrids are fertile. However, reproductive barriers separate the crop complex from the other wild *Cynara* species (Rottenberg and Zohary 1996). Crosses between *C. cardunculus* and *C. syriaca*, *C. algarbiensis*, *C. baetica* and *C. humilis* produce only very few seeds, and the F₁ hybrids are generally sterile. These four wild *Cynara* species are therefore regarded as members of the secondary wild gene pool (GP2) of the globe artichoke and cardoon (Rottenberg and Zohary, 2005). On both morphological (Wiklund, 1992) and cytogenetic (Zohary and Basnizki, 1975) grounds, the closest GP2 member to the cultivated complex is *C. syriaca*. Recently, the monophyly and evolution of *Cynara* has been investigated using ITS (Internal Transcribed Spacer region of nrDNA) sequence data (Robba et al., 2005). Evolutionary studies in five *Cynara* species, based on nucleotide sequence divergences of two internal spacers (ITS1 and ITS2) and an external transcribed spacer (ETS) of the 18S-5.8S-25S rDNA locus, suggest that the crop complex *C. cardunculus* is more differentiated and evolved with respect to the other wild species (Sonnante et al., 2007a).

The wild cardoon is by far the most widely distributed wild *Cynara* taxon, thriving in warm, dry and low altitude environments, but also colonizing man-made habitats such as the edges of cultivated fields and roadsides. It is native to the central and western parts of the Mediterranean basin, extending to the Canary Islands and Madeira. Isolated populations exist further to the east, such as in Cyprus and on the Turkish shore of the Black Sea (Rottenberg and Zohary, 2005). Wild *C. cardunculus* forms are also successful colonizers of extensive areas in the New World, in particular in the South American pampas and more recently in Mexico, California and Australia. The plant is allogamous and seed-propagated (achenes). The majority of seed is shed close to the parent plant, where it germinates rapidly following the first autumn rain, although germination can occur the whole year round under favourable conditions; flowers are usually produced by two year old plants. Like globe artichoke, the fleshy capitula, as well as the petioles and roots, if properly prepared, are edible. Portis et al. (2005a) have used microsatellite and amplified fragment length polymorphism (AFLP) assays to characterise the genetic variation in seven Italian populations of wild cardoon, collected from Sicily and Sardinia, and found that most of the genetic variation was present within, rather than between populations. Furthermore, as a result of geographical isolation, the the Sardinian and Sicilian populations proved highly differentiated, forming two distinct gene-pools.

DNA studies (Lanteri et al., 2004a; Acquadro et al., 2005a), cytogenetic- and isozyme-based (Rottenberg et al., 1996) have all confirmed wild cardoon to be the ancestor of both cultivated *C. cardunculus* forms, which evolved independently

under the influence of distinct anthropogenic selection criteria - globe artichoke for its capitula, and cardoon for its fleshy leaves and stalks. The origin of cardoon and artichoke is presumed to date to the era of Theophrastus (371-287BCE), who described their cultivation in southern Italy and Sicily. The Ancient Greeks and Romans considered artichokes as both a delicacy and an aphrodisiac. In 77CE, the Roman naturalist Pliny the Elder mentioned their use for medicinal purposes, but it was most probably between 800 and 1500CE that the artichoke was domesticated and transformed, presumably in monastery gardens, into the plant which we know today. Although little is known of the process of domestication and subsequent diversification, the significant genetic differentiation between spiny and non-spiny globe artichoke types is suggestive that the two forms evolved in parallel (Lanteri et al., 2004a).

3 Varietal Groups

3.1 Globe artichoke

In Europe, globe artichoke commercial production is, at present, mainly based on the cultivation of perennial, vegetatively propagated clones, which guarantee high yields of marketable product. However a considerable number of new seed propagated cultivars has been developed in recent years, and these are gaining in popularity. The number of vegetatively propagated cultivars grown in the Mediterranean basin and elsewhere is difficult to determine with any accuracy, but is thought to be at least 120. A frequent complication is that a single cultivar can be known by different names in different locations. Around 10-20 clonally propagated cultivars are considered to be of major commercial importance (Basnizki and Zohary, 1994).

Varietal groups are generally identified on the basis of capitulum appearance (Dellacecca et al., 1976; Porceddu et al., 1976; Vanella et al., 1981): (I) the ‘Spinosi’ group has long sharp spines on its bracts and leaves; (II) the ‘Violetti’ group has medium-sized, violet-coloured and less spiny capitula; (III) the ‘Romaneschi’ group has spherical or sub-spherical non-spiny capitula; and (IV) the ‘Catanesi’ group has relatively small, elongated non-spiny capitula. A further classification is based on harvest time: early types can be forced to produce capitula between autumn and spring, if dormant underground shoots used for propagation are watered during summer, whereas late types produce capitula only during spring and early summer (Mauromicale and Ierna, 2000). Early flowering varieties are usually characterized by elongated and small capitula, compared to the globular and larger capitula of the late types (Lopez Anido et al., 1998).

In Italy, globe artichoke production is concentrated in Apulia, Sicily, Sardinia, Tuscany and Lazio. The most commonly grown traditional varieties are the spiny early types ‘Spinoso sardo’ and ‘Spinoso di Palermo’, and the non-spiny types ‘Violetto di Provenza’ and ‘Violetto di Sicilia’ (both early flowering), and ‘Violetto di Toscana’ and ‘Romanesco’ (late). More recently, other clonally propagated varieties have started to make an impact, notably ‘Tema 2000’, a very early, productive cultivar, which produces medium-sized, purple capitula, and ‘Terom’, a

late cultivar producing large, violet capitula, which mature in a well synchronised fashion. In Spain, the most common vegetatively propagated cultivar is 'Blanca de Tudela' (synonym 'Blanca de España'), produced mainly in the south-east coastal region of Murcia and the Navarra; less widespread, but well-prized, is the French cultivar 'Violet de Provence'. In France, globe artichoke varieties are traditionally classified into three groups: (1) 'Brittany artichokes' with large, green, truncated capitula of spherical shape, among which the most commonly grown are 'Camus de Bretagne' and 'Gros Vert de Laon'; (2) 'Midi artichokes', originating from the south of France, with violet leaves and capitula (e.g., 'Violet de Provence', 'Violet de Hyères' and 'Violet du Gapeau'); and (3) secondary cultivars such as 'Blanc Hyerois', which are intermediate in form between 'Camus de Bretagne' and the purple cultivars. In Greece, the major areas of cultivation are the Peloponnese and Crete. The most important cultivars are 'Argos', globe shaped with tightly-closed green bracts, and 'Iodine of Attica', with purple bracts (Bianco, 2005). Outside of Europe, the most commonly grown cultivars are 'Bamafsigi', 'Baladi' and 'Violet de Provence' (the Nile delta region), the Turkish early 'Sakiz' and late 'Bayrampasa', grown, respectively on the Aegean coast and in Marmara (Ercan et al., 2004). In Algeria, Morocco and Tunisia, most cultivars have been introduced from other Mediterranean countries. This is also the case for Argentina, where the vegetatively propagated 'Violet de Provence' represents about 90% of production (Garcia et al., 2004). In the United States, the cultivar 'Green Globe', with large green capitula and thick fleshy scales, grown almost exclusively in coastal California, accounts for about 90% of production; another popular seed propagated cultivar, cultivated mainly in dry areas of California and Arizona, is 'Imperial Star'. In Chile, Peru and China, late seed-propagated cultivars predominate.

The clonally propagated varieties at present in cultivation are highly heterogeneous. DNA marker analysis of the three autochthonous Italian cultivars 'Spinoso sardo', 'Violetto di Sicilia' and 'Spinoso di Palermo' showed that all had a significant element of within-cultivar variation (Lanteri et al., 2001; Portis et al., 2005b). This reflects their multi-clonal composition, which is a direct consequence of the limited selection criteria applied by the farmers. In common practice plants are generally mown at the end of the season, and new propagative material is only collected some months later, without any selection for specific mother plants. An additional source of variation is spontaneous mutation, not necessarily detectable at the phenotypic level, and propagated over time in the absence of a meiotic sieve. Similarly, random amplified polymorphic DNA (RAPD) heterogeneity has been observed in two breeding populations of 'Green Globe' (Tivang et al., 1996) as well as in the cultivars 'Romanesco' and 'Locale di Mola' (Pagnotta et al., 2004; Lotti et al., 2003). Due to the high range of genetic variation found in cultivated populations, Lanteri et al. (2004a) suggested that the term 'varietal type' instead of 'variety' would be more appropriate in order to define the accessions of germplasm at present in cultivation.

Phenotypic chimerae have also been reported within cultivated materials (Pochard et al., 1969). In an AFLP-based genetic uniformity analysis of vegetatively propagated clones from 'Spinoso sardo', Lanteri et al. (2004b) identified three out of 120 individuals showing an electrophoretic profile distinct from that of the parent,

with respect to one or two bands. As these variant patterns were generated from the individual DNAs with at least two AFLP primer combinations, while the profiles from other primer combinations were ‘true to clone’, the authors confirmed that some mother plants must have been chimeric.

3.2 Cardoon

Cardoon production is limited to Spain, Italy and to a limited extent, France. It is usually raised from seed and handled as an annual crop. Plants generated through root division tend to be tough and run rapidly up to flower, while seed-grown plants remain in rosette form and produce more tender leaf ribs. In Spain, the major production area is Navarra, and is mainly based on traditional cultivars, with most of the production (85%) used for canning and freezing. Fresh product is consumed locally from October to March (Macua et al., 2004). The most common varieties for industrial processing are ‘Blanco de Peralta’, a large sized type, with a wide, solid petiole, and ‘Verde de Peralta’, with irregular (wide and tight) but compact petioles, well liked on account of its low fibre content. The varieties destined for the fresh market are the highly productive ‘Verde de Tafalla’, ‘Blanco de Valencia’ (of smaller size and with white hollow petioles), ‘Lleno de España’ (medium to wide petioles), and ‘Rojo de Corella’ (reddish and solid petioles). In Italy, the most commonly grown varieties are ‘Bianco avorio’ (a vigorous type with thick, almost spineless stalks), and ‘Gobbo di Nizza’, an erect plant with thin ribs with some spines, both cultivated mainly in Piedmont (North-Western Italy) and often used uncooked in traditional dishes. In the Emilia region, ‘Bianco pieno migliorato’ has been selected for its solid stalks and leaves, along with ‘Pieno inerme’ and ‘Gigante di Romagna’ (synonym ‘Gigante inerme a foglia intera’), which both have large and very tender stalks completely free of spines.

In France the most commonly grown varieties are ‘Blanc amelioré’, with non-spiny large stems; ‘Plein blanc amelioré puvis’, with broad and thick stalks, sporadic presence of spines and good storability; ‘Vert vaulx velin’ green, spineless and cold resistant and ‘Rouge d’Alger’.

Microsatellite- and AFLP-based analyses have shown a considerable level of within-cultivar variation in both Italian as well as Spanish cultivars, presumably the result of the limited amount of selection that has been applied (Portis et al., 2005c). Seed is commonly mass selected on the basis of the phenotypic conformity of the maternal plant phenotype. Analogous results have been obtained in a RAPD-based study of 17 local Spanish cultivars (Itoiz et al., 2004).

4 Genetic Resources

4.1 Globe artichoke

Italy harbours the richest collection of globe artichoke primary cultivated germplasm and also houses the most abundant *in situ* diversity (Bianco, 1990). Most varietal types are cultivated in limited geographic areas and are identified by vernacular

names. As a result, similar genotypes often carry different names in different localities. Furthermore, distinct varietal types differing from one another due to allelic variation at a small number of genes, can share a largely similar genetic background. Cluster analysis based on the discrimination of eight quantitative characters among 104 accessions was able to identify five major groups sharing similar characters and presumably a similar genome (Elia and Miccolis, 1996). DNA markers have proven to be informative for assessing the extent and distribution of genetic variation in living germplasm collections of globe artichoke, and for identifying suitable strategies for the establishment of core collections. Thus, for example, Sonnante et al. (2002) applied RAPD and AFLP (*ibid.* 2004) markers, but a more complete study was carried out by Lanteri et al. (2004a), who used AFLP markers to investigate patterns of genetic relatedness among 118 accessions, representative of worldwide cultivated germplasm (for three varieties clones of different provenances were included), using two accessions of cultivated cardoon and four of wild cardoon as out-groups. The resulting cluster analysis suggested that the traits selected by man have played an important role in determining variation and differentiation within cultivated artichoke germplasm. Specifically, two major clades could be defined (Figure 3, A and B), each of which was sub-divided into two further clusters. Within the first clade, one cluster (A1) was composed primarily of ‘Catanesi’ types (small elongated capitula), while the other (A2) included the ‘Romaneschi’ types (large spherical or sub-spherical non-spiny capitula and mainly cultivated in central Italy), together with clones of the dominant US variety ‘Green Globe’. The second clade grouped the ‘Spinosi’ and ‘Violetti’ types with medium-small capitula, and all the Turkish accessions lay within one of the sub-clusters (B1). An important purpose of this analysis was to assemble a core collection of globe artichoke, able to take into account the hierarchical structure of the gene pool. The amount of genetic variation between clones of the same variety was, in some cases, higher than that found between varietal types, and thus the lowest JSI (Jaccard Similarity Index) among clones can be taken as a reasonable threshold to identify material sharing the same genetic background.

A more recent study has assessed the level of genetic variation in autochthonous globe artichoke germplasm grown in family gardens in Sicily, at both the morphological and DNA level (Mauro et al., 2007). This collection consisted of 26 morphologically variable types, in particular differing for the number and size of the capitula. AFLP profiling was able to demonstrate that ‘Cimiciusa di Mazzarino’ (many small capitula) is genetically the closest to the wild cardoon, and may therefore represent a transient form of domestication, which is believed to have been achieved in Sicily. The genetic variation in two landraces: ‘Castellamare’ and ‘Campagnano’ of the ‘Romanesco’ type has been assessed in a similar way to identify suitable strategies for preservation (Triofetti Nisini et al., 2007).

4.2 Cardoon

To date, we are aware of only a single attempt to evaluate the genetic variation of cardoon. This study applied microsatellite- and AFLP-profiling to a collection of the most widely grown cultivars in Italy and Spain, plus some local ecotypes, and a few

globe artichoke and wild cardoon entries were used as an out-group (Portis et al., 2005c). The Spanish and Italian cultivars appeared to form two separate gene-pools, but a lack of information regarding the origin of the accessions prevented the establishment of a clear correlation between pedigree and genotype.

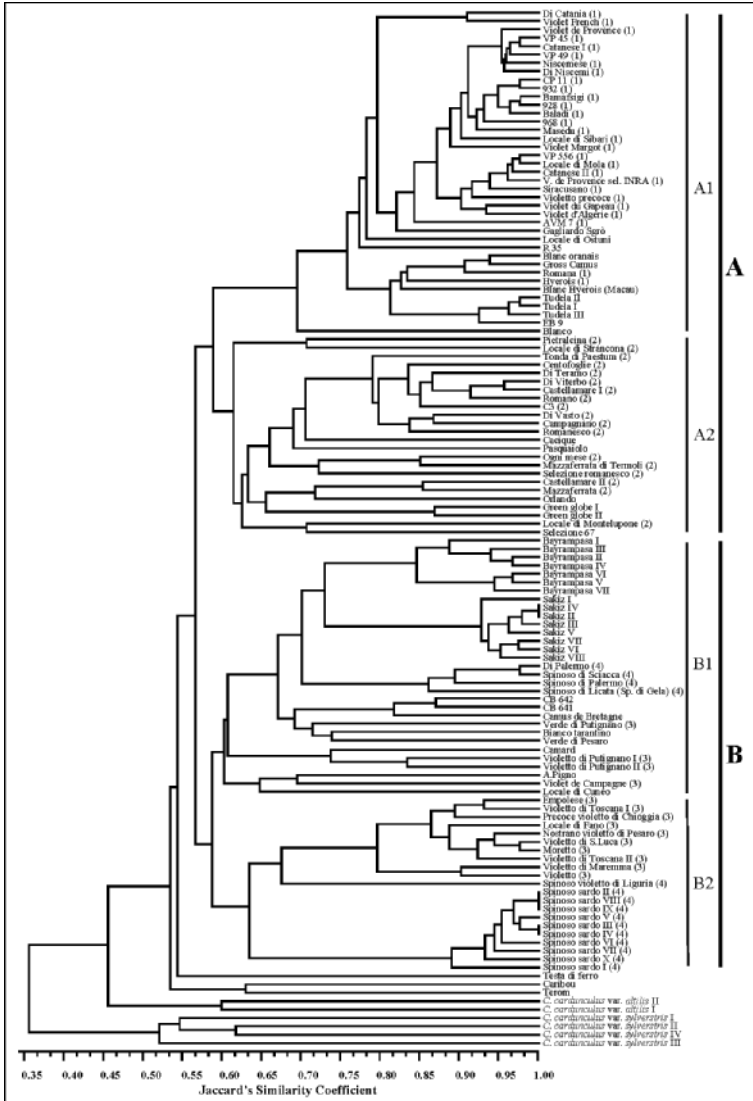


Fig. 3. Dendrogram obtained from UPGMA cluster analysis of AFLP data. (1): ‘Catanesi’ type;(2): ‘Romaneschi’ type; (3): ‘Violetti’ type; (4): ‘Spinosi’ type.

On the whole, a higher degree of genetic differentiation was present in globe artichoke than in the cultivated cardoon. This was not surprising, considering that globe artichoke is cultivated in many localities, but in a fragmentary fashion. As a result, it is likely that a range of selection criteria have been adopted to optimise genotypes both to match individual environments and to suit local tastes.

5 Breeding Methods and Techniques

5.1 Artichoke

5.1.1 Inheritance of Traits

Only few attempts have been made to uncover the inheritance of major capitulum traits (Pécaut, 1993; López Anido et al., 1998; Mauromicale et al., 2000). Most of the morphological and production traits (capitulum size, shape and weight, plant size and branching, peduncle length, earliness and harvest index) are governed by polygenic systems (Porceddu et al., 1976). The thornless bract character has been suggested to be controlled by two genes acting in epistasis, since the segregation between thorn absence and presence from the cross 'Spinoso sardo' x 'Romanesco' was observed to follow a 9:7 ratio (De Pace et al., 1976). However, in a different cross ('Romanesco' x 'Spinoso di Palermo'), its segregation was consistent with a 1:1 ratio, explicable with trait control being exerted by a single gene with two alternative alleles: dominant non-spiny (*Sp*) and recessive spiny (*sp*) (Lanteri et al., 2006). The 'Romanesco' parent was *Spsp* and 'Spinoso di Palermo' *spsp*. Other crosses between non-spiny and either spiny types or wild cardoon (Pochard et al., 1969; Basnizki and Zohary, 1994) have led to the same genetic model, and the *Sp* locus was successfully mapped to linkage group 16 in 'Romanesco', flanked by two microsatellite markers (Lanteri et al., 2006).

There is considerable variation in cultivated germplasm for the intensity of capitulum pigmentation, a character which is also very sensitive to temperature. Nonetheless, varieties are conventionally classified as either violet or green. Pochard et al. (1969) suggested that the genetic basis for anthocyanin pigmentation involves a series of modifiers in addition to one or two major genes. On the basis of segregation in a number of genetic backgrounds, Cravero et al. (2005) have proposed that capitulum colour is genetically determined as follows: *P*- allows anthocyanin production, resulting in purple bracts, while *pp* inhibits anthocyanin production resulting in green bracts; *U*- results in an uneven distribution of anthocyanin pigments encoded by *P*, while *uu* results in an even distribution of pigment in the presence of *P*. Other modifier genes or multiple alleles may also be involved, but the proposed model is sufficient for breeders seeking to enhance colour.

An important quality criterion for the capitulum is tightness, a property important for both the processing and the fresh markets (Macua, 1996). From crosses between soft and fairly compact capitulum types, Dellacecca et al. (1976) showed that the F_1 offspring all had fairly compact capitula; however, when the F_1 hybrids were further crossed with a compact capitulum type, the topcross progeny segregated

4:3:1 for compact: fairly compact: soft capitulum. There also appeared to be a pleiotropic relationship between capitulum form and tightness, with compact capitula being globular or sub-globular because the arrangement of bracts itself contributes to tightness. Cravero et al. (2005) were able to postulate the involvement of two independent loci, *C* and *H*, acting epistatically, such that *C*- genotypes give a compact capitulum, whatever the allelic state of *H*; while *H*- genotypes have tighter capitula in the presence of *cc*.

Lopez Anido et al. (1998) estimated levels of heritability for yield and phenological traits in 23 globe artichoke clones of diverse origin, and which showed extensive genetic variation for these traits, as reported previously (Marzi and Bianco, 1967; Pochard et al., 1969; Foury, 1969). Heritability ranged from 0.94 for capitulum height /diameter ratio, 0.83 for yield, 0.82 for the number of capitula harvested, to 0.48 for the capitulum base diameter. An analysis of variance of the components, broad sense heritabilities and genetic correlations indicated that it should be possible to increase yield and associated characters (i.e., capitula number, weight of secondary capitula, weight of primary capitulum) by 10-30% with a selection intensity of 5%. Lower estimates for broad sense heritabilities were identified in a clonal population of 'Violetto di Sicilia' (Abbate and Noto, 1981), presumably reflecting the lower genetic variation present in this material. The highly significant positive correlation established between capitulum mean diameter and mean weight suggested that selection should favour globular or truncated-conic forms, which have a height/diameter ratio of less than unity (Lopez Anido et al., 1998). The same study established that the weight of the primary capitulum was closely correlated with the base weight and diameter, both of which are important traits for the processing industry; and that many characters were affected by a significant clone x year interaction, as also indicated by Foury (1979) and Mauromicale and Copani (1989).

5.1.2 Breeding Achievements

Globe artichoke ($2n=2x=34$) is cross-pollinated, but (at least in Europe) is generally propagated via suckers, ovoli (underground dormant shoots with a limited root system), or by the division of rooted basal stem portions (De Vos, 1992; Pécaut, 1993). Cross-pollination is promoted by protandry, but self-pollination is not precluded. The stigmatic surface is receptive to pollen for 2-3 days following pollen shedding, and therefore the fertilisation of peripheral florets can be effected by the pollen of the more internal ones, since flowering progresses from the periphery to the centre of the capitulum. Some self-pollination is also possible via pollen transfer between capitula, as each plant produces, on average, four to six asynchronously flowering capitula. Simple strategies of pollen preservation and application allow for the straightforward generation of self-pollinated progeny (Mauromicale and Ierna, 2000), which typically segregate widely. In French and Italian cultivars, the proportion of the first inbred generation (I1) which conformed to the parental phenotype varied from just 2% in 'Violet de Provence' to 8% in 'Romanesco' (Pécaut, 1993); yet repeated selfing, combined with rigorous selection for conformation-to-type, allowed the extraction of true-breeding lines, with a phenotype comparable to that of the parental type (Foury, 1979).

Common breeding targets are the promotion of earliness, yield and capitulum quality, and selection to date has been based largely on intra-clonal variation (Deidda, 1967; Abbate and Noto, 1981; Pécaut, 1993; Mauromicale et al., 2000 Mallica et al., 2004). A list of clones and breeding lines selected in Italy, France and Spain has been presented by Bianco (2005). A few attempts to exploit segregation released by inter-varietal hybridization or selfing have been reported in the literature (Miller, 1975; Scarascia Mugnozza and Pacucci, 1976; Tesi, 1976; Basnizky and Zohary, 1987; 1994). Due to the high level of heterozygosity present in segregating populations, it is normally possible to identify genotypes with valuable agronomic characters, and these selections can then be maintained via vegetative propagation. Some examples of new varieties obtained in this way are ‘Camerys’, ‘Caribou’, ‘Salanquet’, ‘Cacique’, ‘Carlite’ and ‘Terom’ (Mauromicale, 1987). More recently, the green capitulum cultivars ‘Galico’, ‘Castel’, ‘Capitan’, ‘BE15’, ‘Vertu’, ‘Polo’ and the violet ones ‘Velorus’, ‘Salambo’, ‘Satin’, ‘Vialin’ have been released by French INRA stations Plougoum de Saint-Pol de Léon and Avignon-Montfavet. In Italy, a significant area of the ‘Romanesco’ type cultivars ‘Grato 1’, ‘Grato 2’, ‘Etrusco’ and ‘Moro di Corneto’ has been grown (Graifenberg and Giustiniani, 1997; Papalini et al., 1997). Commonly, however, selection within segregating populations derived either by crossing or by selfing of early-flowering types results in late-flowering individuals, which appear to have lost the ability to be forced to produce capitula during the autumn. Thus, for example, Gil and Villa (2004) crossed the Spanish cultivar ‘Tudela’ with the American ‘Imperial Star’ in order to introduce earliness, but after three generations of recurrent selection were not able to obtain any improvement in earliness, which they concluded was under polygenic control. It is also possible that a cycle of sexual reproduction induces a kind of juvenility, with a consequent loss of earliness.

5.2 Cardoon

While globe artichoke is usually vegetatively propagated, cardoon is raised from seed and handled as an annual plant. Seeds are sown in late spring, and the plants over-summer in the vegetative state. Current cultivars have been obtained by mass selection in public or private nurseries. The criteria for cultivar selection are plant size, along with colour and consistency of the pulpy leaves. Commercial cultivars are genetically heterogeneous, as assessed by both RAPD (Itoiz et al., 2004) and AFLP (Portis et al., 2005c) fingerprinting. Further efforts should be carried out to restrict the genetic basis of the material in cultivation and obtain more uniform cultivars.

6 Current Goals of Breeding

The *C. cardunculus* genome is as yet poorly mapped. In order to move to a crossing strategy for breeding, a greater knowledge of artichoke and cardoon genetics will be essential. In particular, it will be advantageous to establish a framework of linkage relationships to allow the identification and localization of genes controlling important yield traits or resistance against pathogens.

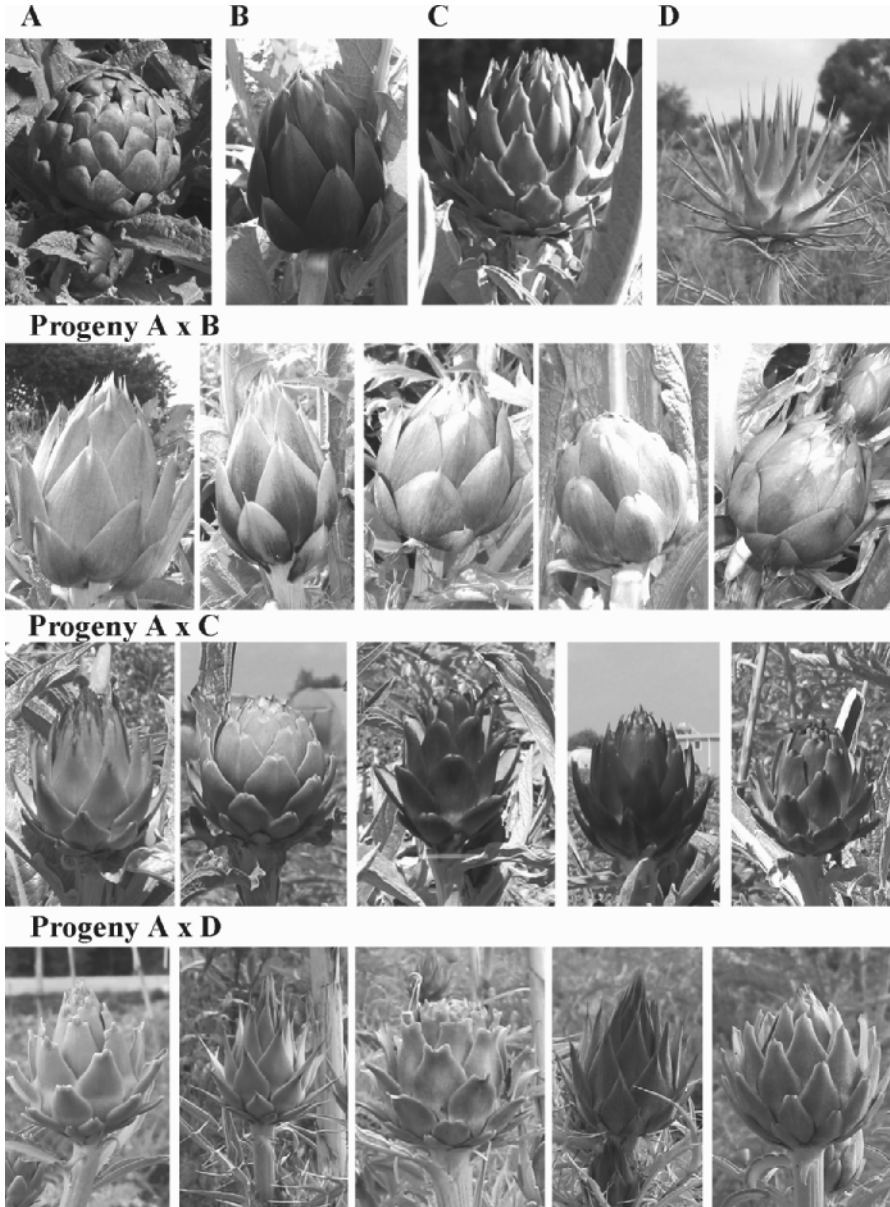


Fig. 4. Head characteristics of the parents crossed for obtaining segregant F_1 population used for the construction of the genetic linkage maps: globe artichoke 'Romanesco clone C3' (A), globe artichoke 'Spinoso di Palermo' (B), cardoon (C) and wild cardoon (D). Examples of morphological variation of the heads observed in the F_1 segregant progenies.

The first genetic maps of globe artichoke, based on a two-way pseudo-testcross strategy, was recently generated by Lanteri et al. (2006). An F₁ population was created by crossing ‘Romanesco clone C3’ (a late-maturing, non-spiny type) with ‘Spinoso di Palermo’ (an early-maturing spiny type), and the progeny were genotyped using a number of marker types (Figure 4). The female map comprised 204 loci, spread over 18 linkage groups and spanned 1330.5cM with a mean marker density of 6.5cM. The equivalent figures for the male parent map were 180 loci, 17 linkage groups, 1239.4cM and 6.9cM. The presence of 78 loci in common to both maps allowed for the alignment of 16 of the linkage groups. The establishment of linkage relationships among such marker loci represents the initial step for the identification of chromosomal regions carrying genes of breeding interest, and their future targeting in breeding programmes via the incorporation of marker-assisted selection. Since globe artichoke is easily vegetatively propagated, the mapping populations are immortalised, and thus can be grown in contrasting environments to investigate genotype x environment interactions, which are known to be important for many commercial traits (see section 5.1.1., above).

Genetic maps are particularly powerful for the dissection of quantitative trait loci (QTL), which underlie the inheritance of many key agronomic characters. The present authors are currently planning the construction of genetic maps based on F₁ populations involving combinations between ‘Romanesco clone C3’ with either cultivated or wild cardoon accessions (Figure 4), as these will allow comparative QTL mapping studies. Wide cross populations of this type are suited for the investigation of the genetic control of quantitative characters in exotic genetic backgrounds. The wild cardoon represents the most straightforward wild resource to exploit for globe artichoke improvement, since it is not genetically isolated from the cultivated gene pool. The other six members of the genus *Cynara* are more problematical, although Rottenberg and Zohary (2005) have succeeded in producing a small number of viable hybrids between four of these and *C. cardunculus*. Possible traits for introduction from exotics include flavour (*C. syriaca*, *C. humilis*), earliness, medicinal properties, dwarfness (*C. cornigera*), white flowers (*C. baetica*, *C. cornigera*) and resistance to pests and diseases, the most damaging being the soil-borne pathogens *Sclerotinia sclerotiorum*, *S. rolfsii*, *Rhizoctonia solani* and *Verticillium daliae*.

7 Integration of New Biotechnologies in Breeding Programmes

7.1 Development and Application of Molecular Markers

To date, molecular marker studies in *C. cardunculus* have been carried out using RAPD (Tivang et al., 1996; Lanteri et al., 2001; Sonnante et al., 2002), AFLP (Lanteri et al., 2004a; 2004b; Sonnante et al., 2004; Portis et al., 2005a; 2005b; 2005c) and Inter-simple sequence repeats (ISSR; Lanteri et al., 2004b). A set of specific microsatellites were developed by Acquadro et al. (2003), using both published and *de novo* acquired (from an enriched genomic library) DNA sequences. Acquadro et al. (2005a) later used the novel “microsatellite amplified library”

(MAL) approach to derive a further set of microsatellite assays. This technique, which represents a combination of AFLP and a primer extension-based enrichment, provides a rapid means to increase the efficiency of microsatellite identification, avoiding the requirement for a hybridization enrichment step (Figure 5). Further *C. cardunculus* microsatellites were then developed using a two-step ‘primer extension’ procedure, based on the microsatellite-AFLP technique (Acquadro et al., 2005b). For this approach, amplicons highly enriched for microsatellite sequences are produced and forward primers directed towards the microsatellite motif are designed from the sequences of bands isolated from the gel profile. Thereafter the opposite microsatellite flanking sequence is isolated via a nested strategy based on a template of restricted-ligated genomic DNA. The extent of the polymorphism uncovered by the total set of 32 microsatellites was explored in a survey of both cultivated and wild accessions. A sequence-specific amplified polymorphism (S-SAP) assay, based on the CYRE-5 Ty1-copia type retrotransposon sequence, was generated by Acquadro et al. (2006) and its effectiveness in assessing genetic variation across 22 *C. cardunculus* accessions, including both cultivated and wild types, was compared to that achievable with AFLP fingerprinting. Finally, 29 of these S-SAP loci were incorporated into the core genetic map, confirming their dispersed distribution across the globe artichoke genome.

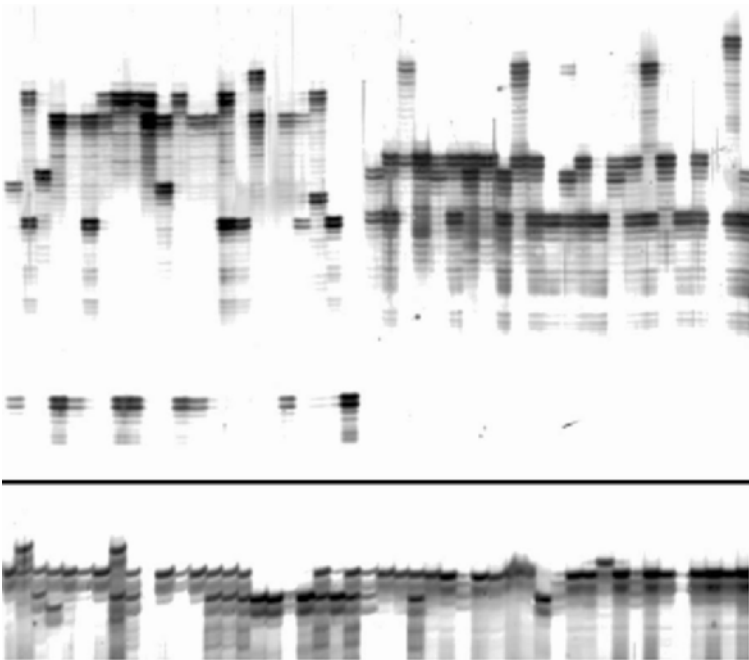


Fig. 5. Polymorphisms observed in a wild cardoon population by applying two microsatellites developed through the novel approach MAL (microsatellite amplified library) at the Di.Va.P.R.A. Plant Genetics and Breeding University of Turin.

7.2 *In vitro* Tissue Culture

Propagation through meristem culture has been widely applied in globe artichoke late types (producing capitula from spring to early summer); the obtained mother virus free plants may represent a source for the production of sanitary controlled propagative material (Barba et al., 2004; Papanice et al., 2004). Plants obtained in this way have shown improved field performance with respect to both qualitative and quantitative traits, and this can compensate for the higher cost of the planting material (Saccardo et al., 2007). On the other hand, the micro-propagation of early-flowering types has frequently produced regenerants which are not true-to-type, specifically in that they often revert to the juvenile stage with a consequent loss of earliness (Cadinu et al., 2004; Frau et al., 2004; Tavazza et al., 2004; Elia et al., 2007). This phenomenon has been ascribed to epigenetic modification of genes involved in the flowering process. A similar epigenetic mechanism may also explain variants arising from vegetative propagation (Pochard et al., 1969; Pécaut and Martin, 1993), such as the 'bull' type described in 'Violet de Provence' and Blanca de Tudela' (Esteva and Martinez, 2004). Recently, Gallitelli et al. (2006) have described how two cycles of meristem culture of the early cultivar 'Brindisino', separated by an *in vitro* *thermotherapy* treatment, allowed for the selection of virus-free plants which retained their earliness under field conditions.

In vitro mutagenesis has been induced by the gamma irradiation of micro-propagated shoots of 'Romanesco clone C3', which were then multiplied for two cycles, rooted and acclimatised (Stamigna et al., 2005). Variants with respect to earliness, bud colour and size and plant height were identified, but further analyses are still needed to validate these putative mutants.

The availability of an efficient protocol for the *in vitro* production of haploid plants and subsequent diploidisation would greatly speed the development of the homozygous material needed for F₁ hybrid breeding. The first reported attempts to culture anthers from five Italian cultivars resulted only in the production of callus (Motzo and Deidda, 1993). Although microspores can now be reproducibly cultured, development beyond the second division has not yet been attained, presumably because of non-optimal culture conditions (Stamigna et al., 2004a; Chatelet et al. 2005). Haploid production via gynogenesis has been also been unsuccessful (Motzo and Deidda, 1993; Babes, 1997), although *in situ* gynogenesis using fertilisation with irradiated pollen has been reported by the INRA station Maraichères de Montfavet (France). However, this method is at present not sufficiently reproducible for general use (Stamigna et al., 2004a).

Finally, *in vitro* tissue culture has been assessed as a route for the preservation of globe artichoke germplasm (Bekheet, 2006). Shoot buds and callus cultures were successfully stored for 15 months at 5°C in the dark, and plantlets regenerated from cultures were successfully adapted to field conditions after simple acclimatization procedures.

7.3 Improvement of Bio-Components of Interest

Modern biotechnology allows the role of genes involved in specific biosynthetic pathways to be defined, and this understanding can potentially increase or modulate the content of key molecules in plant tissue. The chemical composition of globe artichoke leaves and capitula has been extensively studied (Nichiforescu, 1970; Adzet and Puigmacia, 1985; Debenedetti et al., 1993; Slanina et al., 1993; Dranik et al., 1996; Wagenbreth, 1996; Sevcikova et al., 2002; Wang et al., 2003; Di Venere et al., 2007, Mabeau et al., 2007; Melilli et al., 2007). The major phenolic molecules present are the di-caffeoylquinic acids (such as cynarin), which are largely restricted to *Cynara* spp., along with their precursor chlorogenic acid (CGA). The high polyphenol content is the reason why capitula brown after harvest, in a process catalysed by the polyphenol oxidases (oxidoreductases) (Todaro et al., 2006). The anti-oxidative properties of *C. cardunculus* extracts have been tested in rat hepatocytes and shown to possess anti-proliferative and apoptotic effects on cancer cells (Miccadei et al., 2006).

Only pioneer studies have yet been carried out to elucidate biosynthetic pathways of secondary metabolites. A gene sequence(s) encoding a hydroxycinnamoyltransferase (HCT) involved in the synthesis of CGA has recently been isolated in globe artichoke (Comino et al., 2007). *In silico* analysis revealed that this sequence shares homology with one of the five main acyltransferase groups (i.e. anthranilate N-hydroxycinnamoyl/benzoyltransferase). Heterologous expression of the full-length HCT cDNA in *E. coli* demonstrated that the recombinant enzyme efficiently synthesizes p-coumaroyl quinate from p-coumaroyl-CoA and quinic acid, confirming its identity as an hydroxycinnamoyl-CoA: quinate HCT. Variable levels of HCT expression were observed among wild and cultivated forms of *C. cardunculus*. The level of expression was correlated with CGA content, consistent with the predicted involvement of HCT in the biosynthesis of CGA. In an analogous approach, 7et al. (2006b) isolated sequences with high similarity to the PAL (phenylalanine ammonia-lyase) gene family, which encodes enzymes involved in the first step of phenylpropanoid biosynthesis pathway by catalyzing the de-amination of phenylalanine. These sequences differed from one another with respect to intron length, and were differentially expressed in various globe artichoke organs. Root inulin content has been evaluated in wild and cultivated forms of *C. cardunculus*, varying in relation to genotype (higher in wild and cultivated cardoon than in globe artichoke) and stage of plant development (maximum content just before beginning of flower formation) (Melilli and Raccuia, 2007).

8 Globe Artichoke Seed Production

Artichoke seeds (Figure 6) express little dormancy and germinate readily in the range 10-25°C, although higher temperatures do impair germination (Basnizki and Mayer, 1985; Foury 1987). Light inhibits germination, in a cultivar-dependent manner (Basnizki and Mayer, 1985). When stored at room temperature in sealed containers, seeds maintain viability for up to five years (Basnizki and Zohary, 1994).

The effective production of seed of high yielding cultivars is an important objective for artichoke breeding programmes, and could bring about a substantial improvement in both quality and quantity of artichoke production. Seed propagation would allow (i) a reduction in the cost of planting, (ii) a reduction in the spread of pathogens (mainly viruses), (iii) a reduction in fertilizer use and irrigation requirement, since seed-propagated plants develop deeper root systems than do the adventitious roots produced by suckers, and do so in a shorter time, (iv) the selection of varieties whose product can be designed for the industry of transformation (at present predominantly the secondary smaller heads are used in industry), (v) the transformation of artichoke from a perennial to annual crop, which would support crop rotation and thereby limit environmental impact, and (vi) an expansion in the choice of cultivar to the producer. Pioneering studies to obtain seed-propagated cultivars were initiated in France in the 1960's, and began to bear fruit after about twenty years. The first European commercialised seed-propagated cultivars were 'Talpiot', '044', 'Giorgio' and 'Agribas', which produce green capitula between April and May (Basnizky and Zohary, 1987; Mauromicale et al., 1989) and are only slightly sensitive to treatment with gibberellic acid (GA_3), which is commonly widely applied to promote earliness. In the same years, cultivars obtained through mass selection from the cultivar 'Green Globe' were released in USA.

Only limited effort to date has been dedicated to the intra-varietal selection of clones to be used as parents of seed-propagated synthetic varieties. The general and specific combining abilities of several clones have been derived to determine modes of gene action for yield-related and morphological traits (Cravero et al., 2004). In two sets of diallel crosses, with four clones in each, significant differences were established between the mean values of several traits. Most of the variables evaluated were controlled by additive genetic effects, so that recurrent selection should be effective in increasing the mean value of these variables.

F_1 hybrid seed varieties are a highly desirable commodity. First, however, it is necessary to extract uniform, homozygous lines by repeated self-pollination and selection, and to perform a qualitative and quantitative test of F_1 combinations between inbred lines. Globe artichoke suffers, however, from considerable inbreeding depression. Overall plant vigour decreases with increased homozygosity, with deleterious effects observed for leaf area, stem height, the number and size of marketable capitula, the quality and quantity of pollen, and the number of viable seed (Foury, 1979; Pécaut, 1993; Basnizki and Zohary, 1994). Inbreeding depression can appear as early as I_2 , and in some cases, the effects of inbreeding were so marked that it was impossible to continue selfing beyond I_3 or I_4 (Chatelet et al., 2005). Pécaut (1993) found out that, on average, the I_4 generation represents the best compromise between vigour, seed production and homogeneity. By inter-crossing I_3 or I_4 selections, an increase in total average yield of up to 81% was achievable (Basnizki and Zohary, 1994).

The production of F_1 seed propagated varieties would be eased by the identification and incorporation of male sterility (MS) into lines to be used as female parents. A monogenic recessive MS allele, which results in brownish and dry anthers at flowering was identified by Principe (1984), but this source of MS was not fully expressed in other genetic backgrounds (Foury et al., 2005). MS has been identified

in F_2 populations obtained from a cross between a French male sterile clone and both 'Romanesco' and 'Violet de Provence'. This MS generated fully sterile pollen but showed no visible anomalies during tetrad formation, so a post-meiotic mechanism was presumed to be responsible for pollen sterility. It is probably under the control of two recessive genes, since a segregation ratio of 15 male fertile: 1 male sterile was detected in the F_2 of the cross 'MF' x 'MS' (Stamigna et al., 2004b). The first highly vigorous F_1 hybrids on the market were 'H137' and 'H223', producing green capitula with shades of violet from April to May, and lacking sensitivity to GA_3 treatment. More highly GA_3 sensitive F_1 hybrids such as 'Orlando' are now on the market. Following GA_3 treatment, production can begin in November, just a few weeks later than is achieved from clonally propagated early types (Mauromicale and Ierna, 2000).

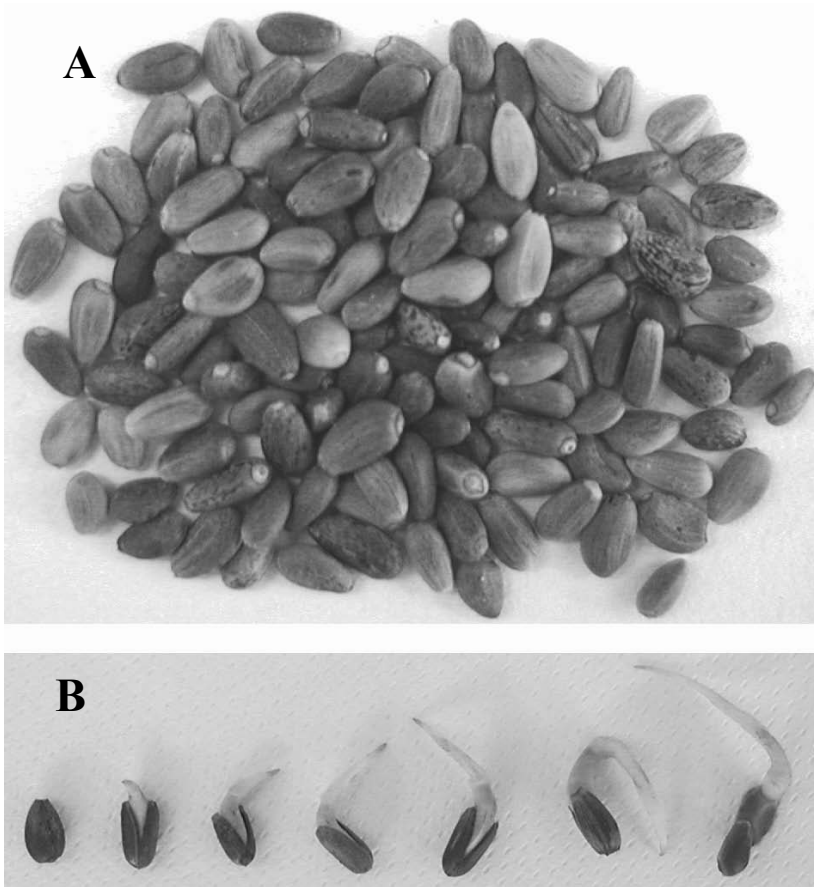


Fig. 6. Dry (A) and germinating (B) seeds of globe artichoke.

Seed-propagated cultivars and F_1 hybrids are now becoming increasingly popular, and account for almost the whole of production in Chile, Peru and China. Numerous trials have been carried out to test their environmental stability. Leskovar et al. (2007) described an evaluation of the seed-propagated cultivars 'Emerald', 'Experimental Red', 'Imperial Star', 'Green Globe' and 'Purple Romagna' in southern Texas, while cultivars 'Concero' and 'Opal', together with some selections commercialized by Nunhems, were tested by Calabrese et al. (2007) in southern Italy and by Miguel et al. (2004a) and Garcia et al. (2004) in southern Spain. No concrete data have yet been obtained regarding the seed propagation of most of the commonly grown early varietal groups. This is mainly because appropriate treatments with GA_3 can only partially advance capitulum production (Miguel et al., 2004b), and in addition, the treatment often induces plant abnormalities such as a reversion to the vegetative state (Condés et al., 2007). The next years are likely to see a large-scale substitution of local landrace types with cultivars selected abroad, with the consequent loss of an important source of local germplasm.

References

- Abbate, V., and Noto, G. 1981. Variabilità ambientale e genotipica in popolazioni siciliane di *Cynara scolymus* ed isolamento di nuovi cloni di Violetto di Sicilia, in: *Atti III Congresso Internazionale Studi sul Carciofo*, ed. Laterza, Bari, pp. 797-807.
- Acquadro, A., Portis, E., and Lanteri, S. 2003. Isolation of microsatellite loci in artichoke (*Cynara cardunculus* L. var. *scolymus* L.), Mol. Ecol. Notes 3:37-39.
- Acquadro, A., Portis, E., Alberini, E., and Lanteri, S. 2005a. M-AFLP based protocol for microsatellite loci isolation in *Cynara cardunculus* L. (*Asteraceae*). Mol. Ecol. Notes 5:272-274.
- Acquadro, A., Portis, E., Lee, D., Donini, P., and Lanteri, S. 2005b. Development and characterisation of microsatellite markers in *Cynara cardunculus* L. Genome 48:217-225.
- Acquadro, A., Portis, E., Moglia, A., Magurno, F., and Lanteri, S. 2006. Retrotransposon based S-SAP as a platform for the analysis of genetic variation and linkage in globe artichoke. Genome 49:1149-1156.
- Adzet, T., and Puigmacia, M. 1985. High-performance liquid chromatography of caffeoylquinic acid derivatives of *Cynara scolymus* L. leaves. J. Chromatogr. 348:447-452.
- Babes, G. 1997. *Impiego di Tecnologie Non Convenzionali Per La Moltiplicazione Del Carciofo*, PhD thesis (X cycle), University of Viterbo, Italy.
- Barba, M., Di Lernia, G., Babes, G., and Citrulli, F. 2004. Produzione e conservazione di germoplasma di carciofo di tipo 'Romanesco' esente da virus. Italus Hortus 11:5-10.
- Barbagallo, R., Chisari, M., Spagna, G., Ierna, A., Patanè, A., Occhipint, A., and Mauromicale, G. 2007. Casein activity expression in flowers of *Cynara cardunculus* spp. Acta Hort. 730:195-199.
- Basnizki, J., and Mayer, M. 1985. Germination in *Cynara* seeds: Effects of light and temperature on the function of the endosperm. Agronomie 5:529-532.
- Basnizki, J., and Zohary, D. 1987. A seed planted cultivar of globe artichoke. HortScience 22:678-679.
- Basnizki, J., and Zohary, D. 1994. Breeding of seed planted artichoke. Plant Breed. Rev. 12:253-269.

- Bekheet, S. 2006. In vitro preservation of globe artichoke germplasm, in: *Proceeding VI International Symposium on Artichoke, Cardoon and Their Wild Relatives*, Lorca-Murcia, Spain, pp. 44.
- Bianco, V. V. 1990. Carciofo (*Cynara scolymus*). In *Orticultura*, ed. V. V. Bianco and F. Rimpini, Patron Editore, Bologna.
- Bianco, V. V. 2005. Present situation and Future Potential of Artichoke in the Mediterranean Basin. *Acta Hort.* 681:39-55.
- Bianco, V. V. 2007. Present and prospects of utilization of fresh and processed artichoke. *Acta Hort.* 730:23-37.
- Brown, J. E., and Rice-Evans, C. A. 1998. Luteolin rich artichoke extract protects low density lipoprotein from oxidation in vitro. *Free. Rad. Res.* 29:247-255.
- Cadinu, M., Repetto, A., Frau, A., Beneventi, S., and Meloni, S. 2004. Influence of The Explant Type on The Phenotypic Changes in Micropropagated Plants of Artichoke. *Acta Hort.* 660:373-380.
- Calabrese, N., De Palma, E., and Damato, G. 2007. Harvest time and yield of artichoke cultivars propagated vegetatively or by seed. *Acta Hort.* 730:345-350.
- Chatelet, P., Stamigna, C., and Thomas, G. 2005. Early development from isolated microspores of *Cynara cardunculus* var. *scolymus* (L.) Fiori, *Acta Hort.* 681:375-380.
- Comino, C., Lanteri, S., Portis, E., Acquadro, A., Romani, A., Hehn, A., Larbat, R., and Bourgaud, F. 2007. Isolation and functional characterization of a cDNA coding a hydroxycinnamoyltransferase involved in phenylpropanoid biosynthesis in *Cynara cardunculus* L. *BMC Plant Biology* 7:14.
- Condés, L. F., Pato, A., and Jiménez, J. 2007. Evaluation of the floral induction and early production of Madrigal F1 artichoke, grown from seed, subjected to different GA3 treatments. *Acta Hort.* 730:171-175.
- Cravero, V. P., Picardi, L. A., and Cointry, E. L. 2005. An approach for understanding the heredity of two quality traits (head color and tightness) in globe artichoke (*Cynara scolymus* L.). *Genet. Mol. Biol.* 28:431-434.
- De Pace, C., Porceddu, E., and Pacucci, G. 1976. Ulteriori risultati di una serie di incroci diallelici nel carciofo, in: *Proceedings II International Congress on Artichoke*, ed. Minerva Medica, Torino, pp. 657-665.
- De Vos, N. E. 1992. Artichoke production in California. *HortTech.* 2:438-444.
- Debenedetti, S. L., Palacios, P. S., Wilson, E. G., and Coussio, J. D. 1993. HPLC analysis of caffeoylquinic acids contents in Argentine medicinal plants. *Acta Hort.* 333:191-199.
- Deidda, M. 1967. Contributo al miglioramento genetico del carciofo, in: *Proceedings I International Congress on Artichoke*, ed. Minerva Medica, Torino, pp. 157-174.
- Dellacecca, V. V., Magnifico, V., Marzi, V., Porceddu, E., and Scarascia Mugnozza, G. T. 1976. Contributo alla conoscenza delle varietà di carciofo coltivate nel mondo, in: *Proceedings II International Congress on Artichoke*, ed. Minerva Medica, Torino, pp. 119-316.
- Di Venere, D., Linsalata, V., Perialice, M., Cardinali, A., Sergio, L., and Crinò, P. 2007. Biochemical characterization of clones from two 'Romanesco' landraces. *Acta Hort.* 730:443-448.
- Drank, L.I., Dolganenko, L. G., Slapke, J., and Thoma, H. 1996. Chemical composition and medical usage of *Cynara scolymus* L. *Rastit. Resur.* 32:98-104.
- Elia, A., and Miccolis, V. 1996. Relationship among 104 artichoke (*Cynara scolymus* L.) accessions using cluster analysis. *Adv. Hort. Sci.* 10:158-162.
- Elia, A., Conversa, G., Montervino, C., Di Brita, S., and Lotti, C. 2007. Micropropagation of the early artichoke cultivar 'Violet de Provence'. *Acta Hort.* 730:127-134.
- Ercan, N., Onus, A. N., Polat, E., Ayar, F., Temirkaynak, M., and Şensoy, A. S. 2004. Determination of optimum GA3 concentrations and awakening irrigation time for globe

- artichoke (*Cynara scolymus* L. cv. Sakýz) grown in mediterranean region of Turkey. Acta Hort. 660:197-200.
- Esteve, J., and Martinez, J. 2004. Evaluation of yield, earliness and head characteristics of bull variant plants in globe artichoke varieties 'Blanca de Tudela' and 'Violet de Provence' at Murcia. Acta Hort. 660:117-121.
- Foti, S., Mauromicale, G., Raccuia, S. A., Fallico, B., Fanella, F., and Maccarone, E. 1999. Possible alternative utilization of *Cynara* spp. I. Biomass, grain yield and chemical composition of grain. Ind. Crop Prod. 10:219-228.
- Foury, C. 1969. Étude de la biologie florale de l'artichaut *Cynara scolymus* L.: Application à la sélection 2. Étude des descendances obtenues en fécondation contrôllée. Ann. Amélior. Plantes 19:23-52.
- Foury, C. 1979. Quelques aspects pratiques de la sélection généalogique de l'artichaut I: presentation, creation de lignées. Ann. Amélior. Plantes 29:383-418.
- Foury, C. 1987. *Quelques Aspects du Développement de l'Artichaut (Cynara scolymus L.) Issu de Semences; Analyse Plus Particuliere de la Floraison en Conditions Naturelles*, These Doctoral, University Orsay, Paris, pp. 189.
- Foury, C., Martin, F., Vaissière, B., Morison, N., and Corre, J. 2005. Avantages et Difficultes de la Creation d'Hybrides F1 d'Artichaut à Semer. Acta Hort. 681:315-322.
- Frau, A., Mallica, G., Baghino, L., Cadinu, M., and Repetto, A. 2004. La micropropagazione del carciofo 'Spinoso sardo': un valido strumento per aumentare la produttività degli impianti. Italus Hortus 11:38-41.
- Gallitelli, D., Papanice, M., Campanale, A., Bottalico, G., and Sumerano, P. 2006. Risanamento nel carciofo 'Brindisino', in: *Atti del Convegno Conclusivo Progetto MIPAF 'Carciofo'*, Roma, pp. 22-24.
- García, S. M., Cointry, E. L., Firpo, I. T., López, F. S., Cravero, V. P., and Asprelli, P. 2004. Influence of sowing dates over seed-grown artichoke production. Acta Hort. 660:387-390.
- Gebhardt, R. 1997. Antioxidative and protective properties of extracts from leaves of artichoke (*Cynara scolymus* L.) against hydroperoxide induced oxidative stress in cultured rat hepatocytes. Toxicol. Appl. Pharmacol. 144:279-286.
- Gebhardt, R. 1998. Inhibition of cholesterol biosynthesis in primary cultured rat hepatocytes by artichoke (*Cynara scolymus* L.) extracts, J. Pharmacol. Exp. Ther. 286:1122-1128.
- Gil, R., and Villa, F. 2004. Breeding for earliness on seed propagated globe artichoke. Acta Hort. 660:35-37.
- González, J., Pérez, F., Fernández, J., Lezaun, J.A., Rodríguez, D., Perea, F., Romero, C., Ochoa, M.J., and García, M. 2004. Study of *Cynara cardunculus* L. lignocellulosic biomass production in dry conditions. Acta Hort. 660:221-228.
- Graifenberg, A., and Giustiniani, L. 1997. Problematiche culturali e valorizzazione del carciofo. Informatore Agrario 27:53-57.
- Itoiz, R., Chocarro, A., and Royo, J. B. 2004. Genetic Variability of Cardoon Populations Evaluated Using RAPD. Acta Hort. 660:249-251.
- Kraft, K. 1997. Artichoke leaf extract. Recent findings reflecting effects on lipid metabolism, liver and gastrointestinal tracts. Phytomedicine 4:369-378.
- Lanteri, S., Di Leo, I., Ledda, L., Mameli, M. G., and Portis, E. 2001. RAPD, variation within and among populations of globe artichoke (*Cynara scolymus* L.), cv. 'Spinoso sardo'. Plant Breed. 120:243-247.
- Lanteri, S., Saba, E., Cadinu, M., Mallica, G. M., Baghino, L., and Portis, E. 2004a. Amplified fragment length polymorphism for genetic diversity assessment in globe artichoke. Theor. Appl. Genet. 108:1534-1544.
- Lanteri, S., Acquadro, A., Saba, E., and Portis, E. 2004b. Molecular fingerprinting and evaluation of genetic distances among selected clones of globe artichoke (*Cynara cardunculus* L. var. *scolymus* L.) 'Spinoso sardo'. J. Hort. Sci. Biotech. 79:863-870.

- Lanteri, S., Acquadro, A., Comino, C., Mauro, R., Mauromicale, G., and Portis, E. 2006. A first linkage map of globe artichoke (*Cynara cardunculus* var. *scolymus* L.) based on AFLP, S-SAP, M-AFLP and microsatellite markers. *Theor. Appl. Genet.* 112:1532-1542.
- Leskovar, D., Goreta, S., Piccinini, G., and Yoo, K. 2007. Strategies for globe artichoke introduction in South Texas. *Acta Hort.* 730:157-163.
- Lopez Anido, F. S., Firpo, I. T., García, S. M., and Cointry, E. L. 1998. Estimation of genetic parameters for yield traits in globe artichoke (*Cynara scolymus* L.). *Euphytica* 103:61-66.
- Lotti, C., De Giovanni, C., Fanizza, G., and Ricciardi, L. 2003. L'analisi AFLP nell'identificazione varietale del carciofo (*Cynara cardunculus* L. var. *scolymus*). *Italus Hortus* 4:246-248.
- Mabeau S., Baty Julienne, C., Anne-Blandine, H., Chodosas, O., Metra, P., and Chesne, C. 2007. Antioxidant activity of artichoke extracts and by-products. *Acta Hort.* 730:491-496.
- Maccarone, E., Fallico, B., Fanella, F., Mauromicale, G., Raccuia, S. A., and Foti, S. 1999. Possible alternative utilization of *Cynara* spp. II. Chemical characterization of their grain oil. *Ind. Crop Prod.* 10: 229-237.
- Macua, J. I., Lahoz, I., Malumbre, A., Garnica, J., Urmeneta, I., and Arrondo, M. A. 2004. Commercial varieties of cardoon in Navarra. *Acta Hort.* 660:215-221.
- Macua, J. I. 1996. Colección de variedades de alcachofa, in: *Actas I Jornadas Técnicas de Alcachofa*, ed. ITGA, Tudela – Navarra, pp. 151-161.
- Mallica, G., Baghino, L., Cadinu, M., and Repetto, A. 2004. Risultati della selezione clonale sulla cultivar di carciofo 'Spinoso sardo'. *Italus Hortus* 11:25-28.
- Martino, V., Caffini, N., Phillipson, J. D., Lappa, A., Tchernitchin, A., Ferraro, G., Debenedelli, S., Schilcher, H., and Acevedo, C. 1999. Identification and characterization of antimicrobial components in leaf extracts of globe artichoke (*Cynara scolymus* L.). *Acta Hort.* 501:111-114.
- Marzi, V., and Bianco, V. V. 1967. Risultati di prove di confronto tra varietà di carciofo di provenienze diverse in Puglia e Lucania, in: *Proceedings I International Congress on Artichoke*, ed. Minerva Medica, Torino, pp. 97-116.
- Matthes, C., and Honermeier, B. 2007. Cultivation of the artichoke as medicinal plant under temperate climate conditions in Germany. *Acta Hort.* 730:483-489.
- Mauro, R., Ierna, A., Portis, E., Lanteri, S., and Mauromicale, G. 2007. Morphological and molecular characterization of autochthonous Sicilian globe artichokes grown in family gardens. *Acta Hort.* 730:113-121.
- Mauromicale, G. 1987. Panorama varietale del carciofo e sua prevedibile evoluzione. *Informatore Agrario* 5:69-75.
- Mauromicale, G., Basnizki, Y., and Cavallaro, V. 1989. Primi risultati sperimentali sulla propagazione del carciofo (*Cynara scolymus* L.) per seme. *Rivista di Agronomia* 23: 417-423.
- Mauromicale, G., and Copani, V. 1989. Caratteristiche biologiche e produzione di cloni diversi di carciofo isolati in popolazioni siciliane di Violetto di Sicilia. *Tecnica Agricola* 41:3-17.
- Mauromicale, G., Morello, N., Santoiemma, G., and Ierna, A. 2000. Nuove varietà per migliorare la cinaricoltura siciliana. *Informatore Agrario* 26:47-51.
- Mauromicale, G., and Ierna, A. 2000. Panorama varietale e miglioramento genetico del carciofo. *Informatore Agrario* 26:39-45.
- Mcdougall, B., King, P. J., Wu, B. W., Hostomsky, Z., Manfred, G., and Robinson, W. E. 1998. Dicafeoylquinic acid and dicafeoyltartaric acid are selective inhibitors of human immunodeficiency virus type 1 integrase. *Antimicrob. Agents Ch.* 42:140-146.
- Melilli, M. G., and Raccuia, S. A. 2007. Inulin and water-soluble –sugars variations in *Cynara* roots during the biological cycle. *Acta Hort.* 730:475-481.
- Melilli, M. G., Trigali, S., Riggi, E., and Raccuia, S. A. 2007. Screening of genetic variability for some phenolic constituents of globe artichoke head. *Acta Hort.* 730:85-91.

- Miccadei, S., Di Venere, D., Cardinali, A., Linsalata, V., Bugianesi, R., Foddai, M. S., Valentini, S., Fraioli, R., and Maiani, G. 2006. Azione antiossidante e apoptotica di frazioni polifenoliche da *Cynara scolymus* in epatociti di ratto e in cellule di epatoma umano, in: *Atti del Convegno Conclusivo Progetto MIPAF 'Carciofo'*, Roma, pp. 104-106.
- Miguel, A., Baixauli, C., Aguilar, J. M., Giner, A., Maroto, J. V., Lòpez, S., and Pascual, B. 2004a. Cultivar Trials of Seed Propagated Artichoke. *Acta Hort.* 660:111-116.
- Miguel, A., Baixauli, C., Aguilar, J. M., Giner, A., Maroto, J. V., Lòpez S., and Pascual, B. 2004b. Gibberellic Acid Concentrations in Seed propagated Artichoke. *Acta Hort.* 660:167-172.
- Miller, T. 1975. New artichoke clones. *New Zealand J. Agr.* 131:33-35.
- Motzo, R., and Deidda, M. 1993. Anther and ovule culture in globe artichoke. *J. Genet. Breed.* 47:263-266.
- Mouzal, L., Aziza, M., Bensiamer-Touati, K., and Hellal-Benateya, A. 2004. Cardoon (*Cynara cardunculus* L.) used as vegetable rennet in an algerina traditional cheese making 'Djben'. *Acta Hort.* 660:207-213.
- Nichiforescu, E. A. 1970. Composition of caffeoylquinic acid derivatives of artichoke (*Cynara scolymus* L.). *Plant Med. Phytother.* 4:56-62.
- Pagnotta, M. A., Cardarelli, M. T., Rey Muñoz, R., Tucci, M., and Saccardo, F. 2004. Assessment of genetic variation in artichoke of 'Romanesco' type by molecular markers. *Acta Hort.* 660:99-104.
- Papalini, P., Cinquanta, A., and Ercolani, R. 1997. Etrusco e Moro di Corneto due nuove cultivar di carciofo. *Informatore Agrario* 27:59-61.
- Papanice, M. A., Campanale, A., Bottalico, G., Sumerano, P., and Gallitelli, G. 2004. Produzione di germoplama risanato di carciofo brindisino. *Italus Hortus* 11:11-15.
- Pécaut, P. 1993. *Globe Artichoke Cynara scolymus L. Genetic Improvements of Vegetable Crops*, ed. Kallo G. and Bergh B.D., Pergamon, Oxford, pp. 737-746.
- Pécaut, P., and Martin, P. 1993. Variation occurring after natural and in vitro multiplication of early Mediterranean cultivars of globe artichoke (*Cynara scolymus* L.). *Agronomie* 13:909-919.
- Perez-Garcia, F., Adzet, T., and Canigual, S. 2000. Activity of artichoke leaf extract on reactive oxygen species in human leukocytes. *Free Radical Res.* 33:661-665.
- Pittler, M. H., and Ernst, E. 1998. Artichoke leaf extract for serum cholesterol reduction. *Perfusion* 11:338-340.
- Pochard, E., Foury, C., and Chambonet, D. 1969. Il miglioramento genetico del carciofo, in: *Proceedings I International Congress on Artichoke*, ed. Minerva Medica, Torino, pp. 117-143.
- Porceddu, E., Dellacecca, V., and Bianco, V. V. 1976. Classificazione numerica di cultivar di carciofo, in: *Proceedings II International Congress on Artichoke*, ed. Minerva Medica, Torino, pp. 1105-1119.
- Portis, E., Acquadro, A., Comino, C., Mauromicale, G., Saba, E., and Lanteri, S. 2005a. Genetic structure of island populations of wild cardoon [*Cynara cardunculus* L. var. *sylvestris* (Lamk) Fiori] detected by AFLPs and SSRs. *Plant Science* 169:199-210.
- Portis, E., Mauromicale, G., Barchi, L., Mauro, R., and Lanteri, S. 2005b. Population structure and genetic variation in autochthonous globe artichoke germplasm from Sicily Island. *Plant Science* 168:1591-1598.
- Portis, E., Barchi, L., Acquadro, A., Macua, J. I., and Lanteri, S. 2005c. Genetic diversity assessment in cultivated cardoon by AFLP (amplified fragment length polymorphism) and microsatellite markers. *Plant Breed.* 124:299-304.
- Principe, J. A. 1984. Male sterility in artichoke. *HortScience* 19:864.
- Quilho, T., Gominho, J., and Pereira, H. 2004. Anatomical characterisation and variability of the thistle *Cynara cardunculus* in view of pulping potential. *Iawa J.* 25:217-230.

- Robba, L., Carine, M. A., Russell, S. J., and Raimondo, F. M. 2005. The monophyly and evolution of *Cynara* L. (*Asteraceae*) sensu lato: evidence from the Internal Transcribed Spacer region of nrDNA. *Plant Syst. Evol.* 253:53-64.
- Rottenberg, A., and Zohary, D. 1996. The wild relatives and the wild ancestry of the cultivated artichoke. *Gen. Res. Crop Evol.* 43:53-58.
- Rottenberg, A., Zohary, D., and Nevo, E. 1996. Isozyme relationships between cultivated artichoke and the wild relatives. *Gen. Res. Crop Evol.* 43:59-62.
- Rottenberg, A., and Zohary, D. 2005. Wild genetic resources of cultivated artichoke. *Acta Hort.* 681:307-311.
- Saccardo, F., Micozzi, F., Di Lernia, G., Piccioni, C., Barba, M., and Pagnotta, M. A. 2007. Virus free artichoke germplasm: quali-quantitative response of globe artichoke. *Acta Hort.* 730:375-379.
- Scarascia Mugnozza, G. T., and Pacucci, G. 1976. Tipi di potenziale valore pratico isolati nell'ambito di un programma per il miglioramento genetico del carciofo, in: *Proceedings II International Congress on Artichoke*, ed. Minerva Medica, Torino, pp. 117-143.
- Sevcikova, P., Glatz, Z., and Slanina, J. 2002. Analysis of artichoke (*Cynara cardunculus* L.) extract by means of micellar electrokinetic capillary chromatography. *Electrophoresis* 23:249-252.
- Slanina, J., Taborska, E. and Musil, P. 1993. Determination of cynarine in the decoctions of the artichoke (*Cynara cardunculus* L.) by the HPLC method. *Cesko-SloV. Farm.* 42: 265-268.
- Slanina, J., Taborska, E., Bochorakova, H., Slaninova, I., Humpa, O., Robinson, W. E., and Schram, H. 2001. New and facile method of preparation of the anti-HIV-1 agent, 1,3-dicafeoylquinic acid. *Tetrahedron Lett.* 42:3383-3385.
- Sonnante, G., De Paolis, A., Lattanzio, V., and Perrino, P. 2002. Genetic variation in wild and cultivated artichoke revealed by RAPD markers. *Gen. Res. Crop Evol.* 49:247-252.
- Sonnante, G., De Paolis, A., and Pignone, D. 2004. Relationships among artichoke cultivars and some related wild taxa based on AFLP markers. *Plant Genet. Res.* 1:125-133.
- Sonnante, G., Carluccio, A. V., Sonnante, G., Vilatersana, L. R., and Pignone, D. 2007a. Variable RDNA regions provide suggestions on artichoke domestication and evolution history. *Acta Hort.* 730:123-125.
- Sonnante, G., De Paolis, A., and Pignone, D. 2007b. Isolation, characterization and expression of PAL gene family in artichoke. *Acta Hort.* 730:81-84.
- Stamigna, C., Crinò, P., Chatelet, P., and Saccardo, F. 2004a. Induction of embryogenesis in isolated microspores of artichoke (*Cynara scolymus* L.). *Acta Hort.* 660:139-14.
- Stamigna, C., Micozzi, F., Pandozy, G., Crinò, P., and Saccardo, F. 2004b. Produzione di ibridi F1 di carciofo mediante impiego di cloni maschiosterili. *Italus Hortus* 11:29-33.
- Stamigna, C., Saccardo, F., Pandozy, G., Ancora, G., and Crinò, P. 2005. *In vitro* mutagenesis of globe artichoke (cv. Romanesco). *Acta Hort.* 681:403-410.
- Tavazza, R., Papacchioli, V., and Ancora, G. 2004. An improved medium for in vitro propagation of globe artichoke (*Cynara scolymus* L.) cv. *Acta Hort.* 660:91-97.
- Tesi, R. 1976. Primi risultati del miglioramento genetico delle varietà toscane di *Cynara cardunculus*, var. *scolymus*, in: *Proceedings II International Congress on Artichoke*, ed. Minerva Medica, Torino, pp. 747-763.
- Tivang, J., Skroch, P. W., Nienhuis, J., and De Vos, N. 1996. Randomly Amplified Polymorphic DNA (RAPD) variation among and within artichoke (*Cynara scolymus* L.) cultivars and breeding populations. *J. Am. Soc. Hort. Sci.* 121:783-788.
- Todaro, A., Iernia, A., Peluso, O., Mauromicale, G., and Spagna, G. 2006. Determination of polyphenol oxidase and studies on browning inactivation in three artichoke cultivars, in: *Proceeding VI International Symposium on Artichoke, Cardoon and their Wild Relatives*, Lorca-Murcia, Spain, pp. 42

- Trionfetti Nisini, P., Crinò, P., Pagnotta, M. A., Gavazza, R., and Ancora, G. 2007. Recovery and characterization of Italian artichoke traditional landraces 'Romanesco' type. *Acta Hort.* 730:101-106.
- Vanella, B., Porceddu, E., and De Pace, C. 1981. Applicazioni di metodi di analisi numerica per il miglioramento genetico del carciofo, in: *Atti III Congresso Int. Di Studi sul Carciofo*, ed. Laterza, Bari, pp. 797-807.
- Wagenbreth, D. 1996. Evaluation of artichoke cultivars for growing and pharmaceutical use. *Beitr. Zuchtungsforsch* 2:400-403.
- Wang, M. F., Simon, J. E., Aviles, I. F., He, K., Zheng, Q. Y., and Tadmor, Y. 2003. Analysis of antioxidative phenolic compounds in artichoke (*Cynara scolymus* L.). *J. Agr. Food Chem.* 51:601-608.
- Wiklund, A. 1992. The genus *Cynara* L. (*Asteraceae-Cardueae*). *Bot. J. Linn. Soc.* 109: 75-123.
- Zohary, D. and Basnizki, J. 1975. The cultivated artichoke *Cynara scolymus*. Its probable wild ancestors. *Econ. Bot.* 29:233-235.