Genome size variation in the *Artemisia arborescens* complex (Asteraceae, Anthemideae) and cultivars.

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Abstract: Different wild Mediterranean populations of *Artemisia arborescens* from diverse locations, representing its geographical distribution, as well as some of its well-known cultivars and some specimens cultivated as ornamentals in gardens, streets, roads and nurseries, were analysed for genome size estimation. Other closely related species endemic to Macaronesia, *A. canariensis*, *A. argentea* and *A. gorgonum*, were also measured, and their nuclear DNA amount has been related to the biogeography of this group of species. Additionally, five populations of the closely related *A. absinthium* were analysed to establish comparisons. Measurements, acquired by flow cytometry, ranged from 8.29 pg to 11.61 pg for 2C values. Statistically significant differences of nuclear DNA amounts with respect to factors such as insularity or domestication have been detected. However, a quite low intraspecific genome size variation has been detected in these species. Furthermore, the study also addressed the possible hybrid origins, and possible misidentifications of some of the supposed cultivars of *A. arborescens*..

Key words: Artemisia arborescens, Artemisia absinthium, Artemisia argentea, Artemisia canariensis, Artemisia gorgonum, C-value, Compositae, cultivar, domestication, flow cytometry, genome size, hybridization, interspecific variation, intraspecific variation, speciation.

Résumé: Plusieurs populations sauvages d'*Artemisia arborescens* provenant de différentes localités représentant toute sa distribution géographique, ainsi que quelques-uns de ses cultivars les plus connus et quelques spécimens cultivés en tant que plantes ornementales dans des jardins, rues, routes et pépinières, ont été analysés pour estimer la taille de leurs génomes. La quantité d'ADN nucléaire a été également mesurée chez d'autres espèces fortement rattachées à celle-ci et endémiques de la région Macaronésienne, *A. canariensis, A. argentea* et *A. gorgonum*, et les résultats ont été mis en rapport avec la biogéographie de ce groupe d'espèces. De plus, cinq populations d'une autre espèce proche de ce complexe, *A. absinthium*, ont été aussi analysées afin d'établir des comparaisons. Les mesures, obtenues par la méthode de cytométrie en flux, vont de 8,29 à 11,61 pg pour les valeurs 2C. Des différences statistiquement significatives de la quantité d'ADN nucléaire ont été détectées, en rapport avec des facteurs tels que l'insularité ou la domestication. Cependant, nous n'avons décelé qu'une assez faible variation intraspécifique de la taille du génome chez ces espèces. L'hypothèse d'une origine hybride et la mauvaise identificaction de quelques cultivars d'*A. arborescens* sont aussi discutées.

Mots clés: Artemisia arborescens, Artemisia absinthium, Artemisia argentea, Artemisia canariensis, Artemisia gorgonum, Compositae, cultivar, cytométrie en flux, domestication, hybridation, spéciation, taille du génome, valeur C, variation interspécifique, variation intraspécifique.

Introduction

The fast-growing shrub *Artemisia arborescens* L., commonly named tree wormwood, silver wormwood or shrubby wormwood is a morphologically variable species, tolerant of a wide range of climates and soil conditions (grows spontaneously or naturalized near human residences) which colonizes a wide geographical range, inhabiting the whole Mediterranean region, across the coastal mainlands and islands. Typically, it is a xerophytic plant of rocks, cliffs and pastures near the sea which forms an upright mound of fine silvery grey leaves with a mild camphor fragrance. According to popular folklore, the plant was spread by Moors and Knights Templar during the times leading up to and including the Crusades and it is frequently found around old fortified sites (Twibell 1992). Medicinal properties, such as antibacterial, anti-inflammatory, antihistamine, anticatarrhal, choleretic and mucolytic are attributed to its essential oils, which are extracted and commercialized (Ikan et al. 1993; Sheppard-Hanger 1995). The medicinal and ornamental uses confer a certain economic interest to this species. This species is phylogenetically close to *Artemisia absinthium* L. (both species belonging to the same subgenus of *Artemisia*, *Absinthium*) with which it shares morphological affinity and ecology, although important chemical differences have been detected between them. Additionally, several cultivars of *A. arborescens*, such as "Brass Band", "Faith Raven", "Huntington", "Little Mice", "Porquerolles" and "Powis Castle", are grown and highly prized in gardens, due to their ever-silver filigree leaves, hardiness and ease of reproduction by cuttings. There is a certain amount of confusion within these cultivars (Twibell 1992, 1994) some of them being considered as simple varieties or forms of *A. arborescens*, whilst others are thought to be hybrids between *A. arborescens* and *A. absinthium* (Twibell 1992). Others are in fact misidentifications of cultivars from other *Artemisia* species which tend to be perpetuated amongst the nursery trade.

There are some species closely related to *A. arborescens* which inhabit Macaronesia, namely *A. argentea* L'Hér., endemic to Madeira (and found in all the islands of the archipelago: Madeira, Porto Santo and Desertas), *A. canariensis* Less. (syn. *A. thuscula)*, endemic to the Canary Islands (where it is found in every island except Lanzarote and Fuerteventura) and *A. gorgonum* Webb, endemic to the Cape Verde archipelago and detected in the islands of São Antão, Santiago and Fogo (Hansen & Sunding 1993). These taxa are considered the vicariant species of *A. arborescens* in Macaronesia, given that *A. arborescens* itself is not present in these islands, and places which it usually colonizes are occupied by the above local endemics. The Açores form an exception however, as no *A. arborescens* or similar species has been found (Hansen & Sunding 1993). *A. arborescens* grows on the North African coasts and it is likely that the presence of these similar taxa in the archipelagos (where they would have differentiated from their ancestor) is due to colonization events from the mainland, as has been described in other genera of Macaronesia (*Cheirolophus*, Garnatje 1995).

Data on genome size can help to clarify many of these aspects. The nuclear DNA content, or C-value, is considered constant within a species; the "C" of C-value stands just for its "constancy" (Swift 1950). Countless studies have been carried out which reveal the existence of a relationship between the nuclear DNA content of a particular species and its ecology, distribution, life cycle, biomass production, resistance, habit or several other features (Bennett 1998; Bennett and Leitch 2005). Amongst these investigations, many workers have focused on re-examining this supposed species-specific constancy, some reporting a considerable intraspecific genome size variation (Rayburn et al. 1985; Ceccarelli et al. 1992; Thibault 1998) and some others supporting intraspecific C-value stability (Bennett et al. 2000; Auckland et al. 2001). Up to now, the extent of intraspecific genome size variation is hotly debated (Greilhuber 2005) and some authors attribute such variation only to methodological errors or taxa misidentification (Greilhuber 1998; Ohri 1998). However, factors like changes in repetitive DNA and retrotransposon activity (Bennetzen

and Kellogg 1997), chromosomal phenomena such as duplications, aneuploidy and the presence of B-chromosomes (Poggio et al. 1998) or even the existence of dimorphic sex chromosomes (Costich et al. 1991), can be a source of variation within a species, amongst other possibilities. It is believed that changes in genome size within a species can be a true indicator of ongoing processes of speciation or genetic divergence (Price 1976; Murray 2005), although it is also true that speciation can take place without any change in nuclear DNA amounts (Bennett and Leitch 2005). Consequently, intraspecific C-value variation is considered in certain cases to be taxonomically significant, because variation in nuclear DNA amounts can be a precedent of reproductive isolation and morphological diversification (Bennett and Leitch 2005). The availability of rapid techniques such as flow cytometry has allowed us the study of a considerable set of populations, geographically distant of the taxa above mentioned (and embracing almost the whole area of distribution of one species, *A. arborescens*). The possibility that nuclear DNA content differs amongst the wild and cultivated populations, cultivars of this species, and the similar Macaronesian taxa, is tested in this study, given the above mentioned features of this core of species. Moreover, we have tried to solve the previously mentioned confusion within the cultivars of *A. arborescens* from the analysis of genome size data.

Materials and methods

Plant material

Young fresh leave tissue was extracted from the studied plants for sample preparation. Some of the seeds of the analysed *A. arborescens* were sampled from Mediterranean populations (including both island and continental representatives), the majority collected from natural sites and one obtained via index seminum; germinated in pots and cultivated under stable conditions in

a greenhouse. The other samples were obtained from adult specimens of *A. arborescens*, some of them also collected from wild populations, some found cultivated in gardens or acquired via plant nurseries or from the NCCPG *Artemisia* Collection. Leaf tissue from the other species *A. canariensis*, *A. argentea*, *A. gorgonum* and *A. vallesiaca* All., as well as from the cultivars, was also obtained from specimens of botanical and nursery gardens or from the NCCPG *Artemisia* Collection. Table 1 shows the populations studied, with an indication of their origin and herbarium voucher information. The NCCPG *Artemisia* Collection holds numerous plants from this genus of known wild or domesticated provenance, which are grown as living specimens. The Collection endeavours to maintain the continuity of living specimens and their genetic variability by vegetative propagation. Collection plants are available for research purposes or for possible future re-introduction.

Seeds of *Pisum sativum* cv 'Express Long', used as internal standard for flow cytometry measurements, were obtained from the Institut des Sciences du Végétal (CNRS, Gif-sur-Yvette, France).

DNA content assessment

The measurements were carried out following the protocols described in a previous study on *Artemisia* L. and related genera (Garcia et al., 2004).

Statistics

Statistical analyses were performed with the Statgraphics Plus 5.0 program (Statistical Graphics Corp., Rockville, Maryland).

Results and discussion

The results for each population are presented in Table 2, together with other data of interest. The analyses were of good quality with a mean HPCV (half peak coefficient of variation) of 2.72%. This is the third study carried on *Artemisia* genome size by our research team (see Torrell & Vallès 2001 and Garcia et al. 2004, for previous results), but the first focused on intraspecific variation and plant domestication within one species. Previously, other works on genome size reported C-values for seven *Artemisia* species (Nagl and Ehrendorfer 1974; Geber and Hasibeder 1980; Greilhuber 1988; Bennett and Smith 1991). According to the categories of genome size (Soltis et al. 2003), the species reported in this study should be considered as intermediate (<3.5 pg to 14.0 pg).

In the present work, nuclear DNA content was determined for the first time for *A. arborescens*. Flow cytometry was used to analyse 35 populations of this species, which represent most of its distribution in the whole Mediterranean basin, as well as some cultivated forms and cultivars (17 wild populations, 8 cultivated in gardens or roads and 10 populations of 6 cultivars of *A. arborescens*). The maximum C-values are those of the population from Mallorca and of one population from Menorca (11.61 pg), and the minimum is the one corresponding to the cultivar "Little Mice" (9.73 pg). Additionally, values obtained for *A. absinthium*, *A. argentea* and *A. canariensis* are consistent with those from previous studies, namely 8.52 pg in Torrell and Vallès (2001) and 9.06 pg in Garcia et al. (2004) for *A. absinthium* (conversely, Nagl & Ehrendorfer in 1974 gave a lower estimate for this species, 7.30 pg), 10.25 pg for *A. argentea* (Greilhuber 1988), and 10.52 pg for *A. thuscula* (synonym of *A. canariensis*) in Torrell and Vallès (2001). In accordance with the data from the plant DNA C-value database (Bennett and Leitch 2004), these are also first C-value estimates for the Cape Verde endemism *A. gorgonum*, and for *A. vallesiaca*, which has been analysed to compare with the cultivar "Little Mice" (this point will be discussed

later). All the species studied share the same chromosome number, 2n=18, according to Kawatani and Ohno (1964), Borgen (1975) and Torrell et al. (1999, 2001), excepting *A. vallesiaca*, with 2n=36 (Kawatani and Ohno 1964).

The rank of variation within *A. arborescens* is: 7.8% within the cultivated, 14% within the studied cultivars and up to 8.8% within the wild populations. This percentage of intraspecific genome size variation within *A. arborescens* is not especially high, and, strictly speaking, the variation detected for this species should be referred to that found for the wild populations. For the populations of *A. absinthium* analysed the intraspecific variation is even lower (6%), and although they come from very different and distant geographical locations, it must be outlined that only five populations have been studied (because the study was not focused in this species).

Compared with other studies, similar variation in genome size within a single species has been detected. For instance, there was a 1.1 fold difference between accessions of *Arabidopsis thaliana* in a study performed by Schmuts et al. (2004). In that work, significant differences ($p<0.05$) were found between all measurements of the five largest diploids and the three accessions with the smallest genome size. Also, in *Silene latifolia*, divergences between male and female individuals from the same population and between geographically separated populations have been reported (Meagher and Costich 1994, 1996; Meagher et al. 2005). On the other hand, many studies show a lower percentage of intraspecific variation. In a study on nuclear DNA amounts of different and geographically isolated populations of *Sesleria albicans*, even though only 1.6% of variation had been detected, the authors found it to be statistically significant (Lysák et al. 2000). In *Hordeum spontaneum*, where genome size for populations representing wide ecological and geographical differences were measured, statistically significant variation up to 5% was found (Turpeinen et al. 1999). This is also the case of *Armeria maritima* in which genome size variation of 7% was related to geographic origin (Vekemans et al. 1996).

Although many of the examples of intraspecific C-value variation have, lately, shown to be artefacts of the measurement methods (Greilhuber 2005; Murray 2005) many reports continue to be published that document genuine intraspecific C-value variation where the appropriate controls and standards have been used (Bennett & Thomas 1991; Reeves et al. 1998; Hall et al. 2000; Moscone et al. 2003). Since it is known that the estimated 100,000 genes that are encoded in an eukaryotic genome make up only approximately 0.12 pg of DNA (Narayan 1998), that the variation takes places, probably, in the non-coding component of the genome which is mainly formed by repetitive DNA (Barakat et al. 1997; Flavell et al. 1997) and that several molecular mechanisms are known which can be responsible for a decrease or an increase in genome size, like the presence of B-chromosomes or transposable elements (more than 60% of some plant genomes are comprised of transposable elements and mostly their defunct remnants, Bennetzen and Kellogg 1997), it is possible that C-value of an species, although fairly constant, admits certain reasonable degree of variation, and thus may not be strictly constant. All things considered, given that measurements were always made with the same internal standard and with the same flow cytometer for each taxon, we believe that the differences detected in this study reflect authentic intraspecific variation.

Effect of domestication, hybrid origin or both?

As has been stated before, we have studied 17 wild populations, 8 populations corresponding to specimens of *A. arborescens* found cultivated in gardens, roads or nurseries and 10 corresponding to 6 different cultivars of this species. Among the cultivars of *A. arborescens* there are certain points of confusion:

1) cultivar "Powis Castle": whilst many nurseries sell this as a "form" of *A. arborescens*, some others suggest that this is a hybrid between *A. arborescens* and *A. absinthium* (see later for details);

2) cultivar "Brass Band": this plant is believed to be identical to "Powis Castle" (Twibell 1992);

3) cultivar "Faith Raven": there are two different plants in circulation under this epithet namely; type 1, which is also thought to be the same as "Powis Castle"; and type 2, which is closer to typical *A. arborescens* (Twibell 1994);

4) cultivar "Porquerolles": a compact form of unknown origin selected from a trial of *A. arborescens* variants at the Porquerolles Botanic Garden near Marseille (France).

5) cultivar "Huntington": another form of intermediate character (or possible hybrid) somewhere between *A. absinthium* and *A. arborescens.* This plant supposedly originated from the Huntington Botanic Garden, San Marino (California, USA) but not is not officially recognised there.

6) cultivar "Little Mice": it is known as "the little brother" of *A. arborescens* in the nursery trade, and sold as a cultivar of this species; however, morphological evidence supports that "Little Mice" is closer to species from subgenus *Seriphidium* of *Artemisia*, particularly to *A. vallesiaca*, than to those from subgenus *Absinthium* that *A. arborescens* belongs to.

All this explained, to reveal the existence of intraspecific genome size variation according to the "degree of domestication" between the different populations of *A. arborescens*, an ANOVA has been performed, and it has resulted in statistically significant differences (P=0.0000) at the 95.0% confidence level. The populations studied have been classified in three categories: wild, cultivated and cultivars. The wild populations have significantly larger genome sizes than the cultivated (approximately 5%), and those larger than the known cultivars (nearly 3%) such as "Powis Castle", "Faith Raven", "Porquerolles" or "Huntington", according to the means comparison test (based on Tukey's Honestly Significant Difference procedure, HSD). Another

ANOVA has been performed excluding the most "doubtful" cultivars of *A. arborescens*, namely "Little Mice" and "Huntington", and also statistically significant differences have been found between the three groups (P=0.0000), although means for the cultivars (10.48) and cultivated (10.78) were not significantly different in the means comparison test. Finally, an additional ANOVA has been done considering as "cultivated" the cultivars "Porquerolles" and "Faith Raven" type 2, given their higher morphological affinity to *A. arborescens* than to the other cultivars, and, again statistically significant differences (P=0.0000) between the groups have been detected, this time being all the means significantly different from each other in the means comparison test.

Given these results, it is conceivable that the process of domestication in *A. arborescens* has lead to a progressive diminution of its genome. In fact, some of the cultivars of this species show a clearly different aspect from the wild *A. arborescens*, with a smaller size or a more compact and silver-grey foliage and a slightly different odour -the essence of these plants has been analysed by Twibell (1992), who found divergences in their vapour profiles. The decrease in genome size of the cultivars with respect to the original species has also been detected in other plants, particularly in crops. Nagato et al. (1981) and Yamamoto and Nagato (1984) reported that in Asian rice and soybean genome size of cultivars was smaller than that of their wild progenitors. Moreover, different varieties of the same crop can have drastic differences in DNA amount, which probably have resulted in evolutionary improvements or in better plant adaptation (e.g. Ceccarelli et al. 1997; Shirasu et al. 2000). For example, different varieties of maize can differ by as much as 42% in their DNA content or different cultivars of chilly pepper differ by 25% (Mukherjee and Sharma 1990; Graham et al. 1994).

On the other hand, a likely hybrid origin of some of the cultivars could explain these differences in genome size, especially those between the wild populations and the cultivars. According to Thomas (1982) the variety "Powis Castle" is reputed to be a hybrid of *A. absinthium* and *A. arborescens*, but contrary to popular opinion it did not originate in the National Trust

garden at Powis Castle (Wales, United Kingdom). The plant was actually taken as a cutting from a plant in a garden (open under the National Gardens Scheme) by Jimmy Hancock (circa 1969-71) who later (1972) became the Head Gardener at Powis Castle (Hancock 1991, private communication). The source of the original plant remains unclear. As Twibell (1992) explains, one possible basis for the "hybrid" theory for the origin of this and other cultivars might derive from their behaviour if severely cut back during the growing season, conditions in which the plants produce simpler greener leaves to maximise photosynthesis. These leaves are fairly similar to those of *A. absinthium*, and the characteristic silver filigree leaves of *A. arborescens* develop subsequently. Additionally, most of these varieties are essentially non-flowering forms (Twibell 1992), another sign of their possible hybrid origin. In this sense, if we calculate the mean value of genome size data for the wild *A. absinthium* (8.60) and for the wild *A. arborescens* (11.28), and also the mean between these figures - which would correspond to, more or less, the expected value for their hybrid –, the resulting number (9.94) is close to the nuclear DNA amount showed by some of the cultivars. On the one hand "Powis Castle", "Faith Raven" type 1 and "Brass Band" show very similar genome sizes (mean nuclear DNA amount of 10.22), a fact that would support them being the same cultivar but with different names; on the other hand, cultivar "Huntington" has a little less, only 9.86 pg. The hypothesis of the hybrid origin of some of these cultivars is supported by these findings, the former ones being closer to *A. arborescens* and the latter closer to *A. absinthium*, on the basis of their genome sizes. Cultivar "Little Mice" has not been included here for the reasons mentioned earlier on; however, its genome size (9.73) is closer to that of *A. vallesiaca* (9.81) than to those of the wild or cultivated *A. arborescens*. This, the fact that no hybrid origin is suspected for "Little Mice", and also the very close morphological affinity with *A. vallesiaca*, lead us to the conclusion that this is not, indeed, a true cultivar of the tree wormwood.

Influence of insularity?

Many of the wild populations analyzed of *A. arborescens* come from Mediterranean islands, and others are from continental origin. It is thought that insular selection pressures can propitiate smaller genome sizes; in this sense, Suda et al. (2003) postulated in a recent work that selection pressures acting on Macaronesian archipelagos favoured small C-values. The same findings have been reported in the insular representatives of the genus *Cheirolophus* (T. Garnatje, S. Garcia and M. A. Canela, unpublished). To test this hypothesis, an ANOVA was performed between the insular and the continental *A. arborescens* (considering both wild and cultivated). Insular species were found to have significantly higher genome sizes than continental species (P=0.0182). However, the fact that all the cultivated species are continental could have biased the analysis, being this difference a consequence of the domestication process rather than a genuine difference between island and continental populations. Therefore, the ANOVA was carried on only considering the wild populations, and the difference is not significant anymore (P=0.9799). In other words, island and continental populations in terms of genome size are quite equal, and this reinforces the notion that genome size is fairly constant within a species.

Evidence of speciation?

Comparing the mean genome size of the wild populations of *A. arborescens* with those of the species which occupy the same ecogeographical placement in some of the islands of Macaronesia (i.e. *A. argentea* in Madeira, *A. canariensis* in Canary Islands and *A. gorgonum* in Cape Verde) it is noticeable that these latter have considerably less nuclear DNA than the former. As has been previously stated, it is quite possible that these Macaronesian species are the vicariants of *A. arborescens*, because all of them have high morphological affinity and the same chromosome number, and also occupy the same ecological niche which *A. arborescens* would fill if present in Macaronesia. In other words, these Macaronesian taxa could have undergone a process of speciation, which probably has been reflected in a decrease in genome size due to stronger selective constraints in the Atlantic islands compared to the Mediterranean Islands, where

the studied populations of *A. arborescens* grow. As previously stated, insular selection pressures in Macaronesia have favoured small C-values. Because none of Macaronesian islands were part of a continent, the native plants probably reached the island by long-distance dispersal. Given that *A. arborescens* is also present in the North African coast (three of the populations studied come from North Africa, two from Algeria and one from Morocco), it is likely that seeds from these continental populations propagated to Macaronesian archipelagos, and subsequently differentiated into separated species, as an adaptive response to each of the various islands' environments (this mechanism has also been described for other Macaronesian taxa, Garnatje 1995). Actually, it has been shown that evolutive phenomena of speciation and adaptive radiation occur faster in insular ecosystems, particularly in oceanic islands, than in continents (García-Talavera 1999). On the other hand, the fact that none of these taxa is present in Lanzarote or in Fuerteventura, which are the closest islands to the North African coast, is probably due to their high volcanic activity in the past (which could have caused massive species extinction) and the particularly arid climatic conditions of these two islands.

However, speciation may occur without any detectable change in C-value (Bennett and Leitch 2005), but there are available examples of considerable intraspecific genome size variation that, together with clear morphological and ecological differences can lead to species split (the case of *Lachnagrostis littoralis*, Murray 2005). Amidst the known mechanisms which can lead to a genome size decrease there are the processes of unequal intrastrand homologous recombination, the illegitimate recombination or the loss of DNA during the repair of double stranded breaks; the available studies suggest that deletional mechanisms may play a more prominent role in genome size evolution than previously thought (Bennett and Leitch 2005).

Concluding remarks

Although moderately low, genuine intraspecific variation has been found within the different populations of *A. arborescens* and *A. absinthium* analysed. It has been clearly shown that genome size varies greatly across plant species, and both increases and decreases can be related to evolutionary events (Bennett and Leitch 2005; Cullis 2005). While in a certain group of taxa evolution can point to a gain in total nuclear DNA amount, in some others the tendency is to reduce it, and consequently it is extremely difficult if not impossible to establish a general pattern of change in genome size. As Bennett and Leitch stated (2005) variation in DNA amount between species begins with changes within species, which implicitly recognizes that C-values are fairly constant but allow certain degree of intraspecific variation. If we accept the existence of a certain degree of intraspecific variation, and that genome size diversification is an important process during speciation in plants (Greilhuber 1998; Soltis et al. 2003) we must wonder what percentage of variation is "acceptable" within a single species. There is a similar dilemma in the issue of establishing a "sufficient amount" of genetic differentiation that can be associated with speciation, because there are studies showing that extensive amounts of genetic differentiation are related with speciation, although not all the variability observed can be directly related to the speciation process (Hancock 2003). Ultimately, what is questioned here is the concept of a species. Additional comparative studies on genome size variation within a species in relationship to morphological, environmental or ecogeographical differences between its populations are needed in order to establish such a criterion.

Additionally, this study has also helped to clarify the confusion existing within the different cultivars of *A. arborescens* using data on nuclear DNA amounts. From our findings, we conclude that cultivars "Powis Castle" and "Huntington" are of likely hybrid origin between *A. arborescens* and *A. absinthium*; cultivars "Brass Band" and "Faith Raven" type 1 are the same as "Powis

Castle"; and cultivars "Porquerolles" and "Faith Raven" type 2 are simply cultivated forms of *A. arborescens*. Finally "Little Mice" is not a cultivar of *A. arborescens*: from its morphological appearance and genome size it is closer to species from subgenus *Seriphidium.*

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References

Auckland, L.D., Johnston, J.S., Price, H.J., and Bridgewater, F. E. 2001. Stability of nuclear DNA content among divergent and isolated populations of Fraser fir. Can. J. Bot. **79:** 1375-1378.

Barakat, A., Carels, N., and Bernardi, G. 1997. The distribution of genes in the genomes of the Gramineae. Proc. Nat. Acad. Sci. USA **94**: 6857-6861.

Bennet, M.D. 1998. Plant genome values: how much do we know? Proc. Natl. Acad. Sci. U.S.A. **95:** 2011-2013.

Bennett, M.D., and Leitch, I.J. 2004. Plant DNA C-values Database (release 3.0, December 2004). Available from http://www.rbgkew.org.uk/cval/homepage.html.

Bennett, M.D., and Leitch, I.J. 2005. Genome size evolution in plants. *In* The evolution of the genome. *Edited by* T.R. Gregory. Elsevier Academic Press, USA. pp. 90-164.

Bennett, M.D., and Smith, J.B. 1991. Nuclear DNA amounts in angiosperms. Ann. Bot. (London) **76**: 113-176.

Bennett, M.D., Johnston, S., Hodnett G.L., and Price, H.J. 2000. *Allium cepa* L. cultivars from four continents compared by flow cytometry show nuclear DNA constancy. Ann. Bot. (London) **85**: 351-357.

Bennett S.T., and Thomas, S.M. 1991. Karyological analysis and genome size in *Milium* (Gramineae) with special reference to polyploidy and chromosomal evolution. Genome **34:** 868–878.

Bennetzen, J.L., and Kellogg, E.A. 1997. Do plants have a one-way ticket to genomic obesity? Pl. Cell **9(9):** 1509-1514.

Borgen, L. 1975. Chromosome numbers of vascular plants from Macaronesia. Norwegian J. Bot. **22:** 71-76.

Ceccarelli, M., Falistocco, E., and Cionini, P.G. 1992. Variation of genome size and organisation within hexaploid *Festuca arundinacea*. Theor. Appl. Gen. **83:** 273-278.

Ceccarelli, M., Girodani, T., Natali, L., Cavallini, A., and Cionini, PG. 1997. Genome plasticity during seed germination in *Festuca arundinacea*. Theor. Appl. Gen. **94:** 309-315.

Costich, D.E., Meagher, T.R., and Yurkow, E.J. 1991. A rapid means of sex identification in *Silene latifolia* by use of flow cytometry. Plant. Mol. Biol. Report. **9:** 359-370.

Cullis, A.C. 2005. Mechanisms and Control of Rapid Genomic Changes in Flax. Ann. Bot. **95(1):** 201-206.

Flavell, A.J., Pearce, S.R., Heslop-Harrison, J.S., and Kumar, A. 1997. The evolution of *Ty1-copia* group retrotransposons in eukaryote genomes. Genetica **100:** 185 195.

Garcia, S., Sanz, M., Garnatje, T., Kreitschitz, A., McArthur, E.D., and Vallès, J. 2004. Variation of DNA amount of 47 populations of the subtribe Artemisiinae and related taxa (Asteraceae, Anthemideae): karyological, ecological and systematic implications. Genome **47:** 1004-1014.

García-Talavera, F. 1999 La Macaronesia. Consideraciones geológicas, biogeográficas y palaeoecológicas. *In* Ecología y cultura en Canarias. *Edited by* J. M. Fernández-Palacios, J. J. Bacallado, and J. A. Belmonte [eds.]. Museo de la Ciencia, Cabildo Insular de Tenerife, Santa Cruz de Tenerife. pp. 39– 63.

Garnatje T. 1995. Relacions entre el polimorfisme enzimàtic i alguns aspectes de l'ecologia i de l'evolució en el gènere *Cheirolophus* Cass. Universitat Autònoma de Barcelona. PhD thesis.

Geber, G., and Hasibeder, G. 1980. Cytophotometrische Bestimmung von DNA. Mengen: vergleich einer neuen DAPI-Fluoreszenz Methode mit Feulgen-Absortionsphotometrie. Microsc. Acta, **4(suppl.):** 31-35.

Graham, M.J., Nickell, C.D., and Rayburn, A.L. 1994. Relationship between genome size and maturity group in soybean. Theor. Appl. Genet. **88:** 429-432.

Greilhuber, J. 1988. "Self-tanning"- a new and important source of stoichiometric error in cytophotometric determination of nuclear DNA content in plants. Pl. Syst. Evol. **158:** 87-96.

Greilhuber, J. 1998. Intraspecific variation in genome size: a critical reassessment. Ann. Bot. **82 (suppl. A):** 27–35.

Greilhuber, J. 2005. Intraspecific variation in genome size in angiosperms: identifying its existence. Ann. Bot. **95:** 91-98.

Hancock, JF. 2003. Plant Evolution and the Origin of Crop Species, 2nd Edition. CABI Publishing, Cambridge, MA, USA.

Hall, S.E., Dvorak, W.S., Johnston, J.S., Price, H.J., and Williams, C.G. 2000. Flow cytometric analysis of DNA content for tropical and temperate new world pines. Ann. Bot. **86:** 1081–1086.

Hansen, A, and Sunding, P. 1993. Flora of Macaronesia. Check list of vascular plants. 4th Ed. Sommerfeldia 17. Oslo, Norway.

Ikan, R., Deans, S.G., and Vavid, U. 1993. Antibacterial, antifungal and antioxidant properties of five essential oils from Israel. Program Abstracts, 24th International Symposium of Essential Oils.

Kawatani, T., and Ohno, T. 1964. Chromosome numbers in *Artemisia*. Bull. Nat. Inst. Hyg. Sci. Jap. **82:** 183-193.

Lysák, M.A., Rostková, A., Dixon, J.M., Rossi, G., and Doležel, J. 2000. Limited genome size variation in *Sesleria albicans*. Ann. Bot. **86:** 399-403.

Meagher, T.R., and Costich, D.E. 1994. Sexual dimorphism in nuclear DNA content and floral morphology in populations of *Silene latifolia* (Caryophyllaceae). Am. J. Bot. **81:** 1198–1204.

Meagher, T.R, and Costich, D.E. 1996. Nuclear DNA content and floral evolution in *Silene latifolia*. Proc. Roy. Soc. Lond. (B) Biol. Sci. **263:** 1455–1460.

Meagher, T.R., Gillies, A.C.M., and Costich, D.E. 2005. Genome size, quantitiative genetics, and the genomic basis for flower size evolution in *Silene latifolia. Ann. Bot.* **95:** 247-254.

Moscone, E.A., Baryani, M., Ebert, I., Greilhuber, J., Ehrendorfer, F., and Hunziker, A.T. 2003. Analysis of nuclear DNA content in *Capsicum* (Solanaceae) by flow cytometry and Feulgen densitometry. Ann. Bot. **92:** 21–29.

Mukherjee, S., and Sharma, K.A. 1990. Intraspecific variation of nuclear DNA in *Capsicum annuum* L. Proc. Ind. Acad. Sci. **100:** 1-6.

Murray, B.G. 2005. When does intraspecific C-value variation become taxonomically significant? Ann. Bot. **95:** 119-125.

Nagl, W., and Ehrendorfer, F. 1974. DNA content, heterochromatin, mitotic index, and growth in perennial and annual Anthemideae (Asteraceae). Pl. Syst. Evol. **123:** 737-740.

Nagato, Y., Yamamoto, K., and Yamashita, H. 1981. Variation of DNA content in Asian rice. Jap. J. Gen. **56:** 483-493.

Narayan, R.K.J. 1998. The role of genomic constraints upon evolutionary changes in genome size and chromosome organization. Ann. Bot. **82 (Suppl. A):** 57-66.

Ohri, D. 1998. Genome size variation and plant systematics. Ann. Bot. **82:** 75-83.

Poggio, L., Rosato, M., Chiavarino, A.M., and Naranjo, C.A. 1998. Genome size and environmental correlations in maize (*Zea mays* ssp. *mays,* Poaceae). Ann. Bot. **82 (Suppl. A):** 107-117.

Price, H.J. 1976. Evolution of DNA content in higher plants. Bot. Rev. **42:** 27–52.

Rayburn, A.L., Price, H.J., Smith, J.D., and Gold, J.R. 1985. C-band heterochromatin and DNA content in *Zea mays*. Am. J. Bot. 72: 1610-1617.

Reeves, G., Francis, D., Davies, M.S., Rogers, H.J., and Hodkinson, T. 1998. Genome size is negatively correlated with altitude in natural populations of *Dactylis glomerata.* Ann. Bot. **82 (Suppl. A):** 99–105.

Schmuts, H., Meister, A., Horres, R., and Bachmann, K. 2004. Genome size variation among accessions of *Arabidopsis thaliana*. Ann. Bot. **93:** 1-5.

Sheppard-Hanger S. 1995. The aromatherapy practitioner reference manual. Atlantic Institute of Aromatherapy. Tampa, Florida.

Shirasu, K., Schulman, A.H., Lahaye, T., and Schulze-Lefert, P. 2000. A contiguous 66 kb barley DNA sequence provides evidence for reversible genome expansion. Gen. Res. **10:** 908-915.

Soltis, D.E., Soltis, P.S., Bennett, M.D., and Leitch, I.J. 2003. Evolution of genome size in the angiosperms. Am. J. Bot. **90:** 1596–1603.

Swift, H. 1950. The constancy of deoxyribose nucleic acid in plant nuclei. Proc. Natl. Acad. Sci. U.S.A. **36:** 643-654.

Suda, J., Kyncl, T., and Freiová, R. 2003. Nuclear DNA amounts in Macaronesian Angiosperms. Ann. Bot. **92:** 153-164.

Thibault, J. 1998. Nuclear DNA amount in pure species and hybrid willows (Salix): a flow cytometric investigation. Can. J. Bot. **76:** 157-165.

Thomas, G.S. 1982. Perennial garden plants or the modern florilegium. p. 69. J. M. Dent & Sons Ltd. G. B., London.

Torrell, M., and Vallès, J. 2001. Genome size in 21 *Artemisia* L. species (Asteraceae, Anthemideae): Systematic, evolutionary and ecological implications. Genome **44:** 231-238.

Turpeinen, T., Kulmala, J., and Nevo, E. 1999. Genome size variation in *Hordeum spontaneum* populations. Genome **42:** 1094-1099.

Torrell, M., Garcia-Jacas, N., Susana, A., and Vallès, J. 1999. Phylogeny in *Artemisia* (*Asteraceae, Anthemideae*) inferred from nuclear ribosomal DNA (ITS) sequences. Taxon **48:** 721-736.

Torrell, M., Vallès, J., Garcia-Jacas, N., Mozaffarian, V., and Gabrielian, N. 2001. New or rare chromosome counts in the genus *Artemisia* L. (Asteraceae, Anthemideae) from Armenia and Iran. Bot. J. Linn. Soc. **135:** 51-60.

Twibell, J.D. 1992. Plant identification from vapour analysis, a short study of *Artemisia arborescens* varieties. Plantsman **14(3):** 184-190.

Twibell, J.D. 1994. Artemisia 'Faith Raven'. Plantsman **15(4)**: 255-258.

Vekemans, X., Lefèbvre, C., Coulaud, J., Blaise, S., Gruber, W., Siljak-Yakovlev, S., and Brown, S.C. 1996. Variation in nuclear DNA content at the species level in *Armeria maritima*. Hereditas **124:** 237-242.

Yamamoto, K., and Nagato, Y. 1984. Variation of DNA content in the genus *Glycine*. Jap. J. Breed. **38:** 470-473.

Table 1. Provenance of the populations of *Artemisia* studied.

Taxa	Locality, collector(s), and date	Herbarium voucher[a]
A. arborescens	**(Populations cultivated in gardens, streets, roads or nurseries)**	
	France, París. Purchased in a market. J. Vallès, II-2005.	BCN 30415
	France, Portús. Cultivated as ornamental in a garden. J. Vallès, 28-III-2005.	BCN 30420
	Morocco, Rif. Cultivated in a private garden. O. Hidalgo & A. Romo, 20-VI-2005.	BCN 30560
	Spain, Catalonia, Barcelona, Montjuïc. Historical Botanic Garden. M. Veny, III-2005.	BCN 30403
	Spain, Catalonia, Barcelona, Montjuïc. Cultivated as ornamental in the nursery "Tres Pins". T. Garnatje, III-2005.	BCN 30423
	Spain, Catalonia, La Garrotxa, Besalú. Planted in a roundabout of the road from Girona to Besalú, near the "Pont Vell". J. Vallès, 26-VI-2005.	BCN 30561
	Spain, Madrid. Botanic Garden. J. Vallès, 16-V-2005.	BCN 30502
	Spain, València. Purchased in the nursery "Pro Agri". J. Vallès, 1-VI-2005	BCN 30540
A. arborescens	**(Wild populations)**	
	Algeria. National Park of Gouraya, Bejaia. J. Vallès, 11-X-2004.	BCN 29895
	Algeria. Botanic Garden Essais Hamma. From a wild locality of Algeria. J. Vallès, 21-IX-2004.	BCN 29950
	France, Corsic, Bonifaziu. Over the walls of the ancient Citadelle. M. Bosch & M. R. Orellana, 9-V-2005.	BCN 30516

France, Porquerolles. Maritime sands. From the Index Seminum of the Museum of Natural History of Paris, III-2004.	BCN 29547
Greece, Corfú, Armenodes. NCCPG *Artemisia.* L. Chilton, 1995.	NCCPG 1995/76
Greece, Crete. Kalyves beach. GR-128. T. Garnatje & J. Luque, 15. VIII.2002.	BCN 27513
Greece, Rhodes. Mount Filerimos. NCCPG *Artemisia.* B.Tickner, IV-1993.	NCCPG 1994/68
Italy, Sardinia, Alguero. Wayout urban nucleous by Fertilia road margins, T. Garnatje & J. Vallès, 7.XII.1999.	BCN 25512
Italy, Sicily, Sciacca. Gole della Tardara, river Carboi. V. Ilardi, G. Domina & C. Blanché. 10-V-2005.	BCN 30494
Spain, Alacant, La Encina. Near "La Casa de los Corrales". T. Garnatje & R. Vilatersana (v-456), 23-V-2005.	BCN 30513
Spain, Balearic Islands, Formentera. T. Garnatje & R. Vilatersana, 18-IV-04.	BCN 30470
Spain, Balearic Islands, Formentera. T. Garnatje & R. Vilatersana, 18-IV-04.	BCN 30471
Spain, Balearic Islands, Mallorca. S'Alqueria Blanca, "Santuari de la Consolació", enclosure margins, at 200 m. J. Vicens. 24.VI.1997.	BCN 22321
Spain, Balearic Islands, Menorca, Maó. Cliffs near the harbour, 2 km from Es Castell. A. Gómez, M.A. Ribera, J.A. Seoane & J. Vallès, 19-I-1997.	BCN 22115
Spain, Menorca. Cliffs at Binibeca. NCCPG *Artemisia.* J. Twibell, VIII. 1995.	NCCPG 1995/74
Spain, Tarragona. Road from Roquetes to Alfara de Carles (TV3422), km 8 (near the canal), enclosure of a house.T. Garnatje & R. Vilatersana, 26-V-2005.	BCN 30514
Turkey, Hatay, Samandag, Çevlik, Kral Mezarlarý Mevkii. Rocky cliffs, at 10m over the sea level. Fadime Gumusboga, 7-VI-2005.	BCN 30550

Cultivars of *A. arborescens*

"Brass Band"	England, United Kingdom, NCCPG *Artemisia*. Geoff Hamilton 1989.	NCCPG 1989/01
"Faith Raven" type 1*	England, United Kingdom, NCCPG *Artemisia*. Faith Raven, 1990.	NCCPG 1990/03
"Faith Raven" type 1*	England, United Kingdom, NCCPG *Artemisia*. Ex John or Faith Raven (1969). Via J. Goulsbra, 1993.	NCCPG 1993/04
"Faith Raven" type 2*	England, United Kingdom, NCCPG *Artemisia*. Ex John or Faith Raven (circa 1970). Via Dr. Jamison , XII-1993.	NCCPG 1993/05
"Huntington"	England, United Kingdom, from Heronswood Nursery Seattle USA. Via N Pope (Hadspen House Nursery, UK) and R Mort, 1999.	NCCPG 1999/01
"Little Mice"	England, United Kingdom, NCCPG Artemisia 2005/29 from Chilton Quality Plants nursery. J. Twibell, 2005.	NCCPG 2005/29
"Little Mice"	France, Theix. Purchased in the nursery "Le Clos d'Armoise". S. Garcia, V-2005.	BCN 30513
"Porquerolles"	France. Conservatoire Botanique de Porquerolles, near Hyères, Form of unknown origin selected from trial stock of arborescens variants. NCCPG *Artemisia*. J. Simmons (RBG Kew) 28-X-1986.	NCCPG 1990/05
"Powis Castle"	England, United Kingdom. Blooms of Bressingham. NCCPG *Artemisia* J. Twibell, 1988.	NCCPG 1988/08
"Powis Castle"	Spain, Catalonia, Barcelona, Sant Andreu de Llavaneres. Purchased in the nursery "Sala-Graupera", S. Garcia & J. Vallès, III-2005.	BCN 30425

Other related species and cultivars

A. absinthium	Armenia. NCCPG *Artemisia*. J. Vallès, 1997.	NCCPG 1997/54
A. absinthium	Iran. Seed from Teheran Botanic Garden. J. Twibell, 1994.	NCCPG 1994/54
A. absinthium	Spain, Andalusia. Sierra Nevada. Near Alburgue and northern road barrier Picos de Valetta. NCCPG *Artemisia*. J. Twibell, X1-2000.	NCCPG 2000/84
A. absinthium	Spain, Catalonia, Girona, Maçanet de Cabrenys. J. Vallès, IV-2005.	BCN 30476

A. absinthium	France, Mèze. Purchased in the nursery "Pépinière Filippi". S. Garcia, VI-2005.	BCN 30567
A. argentea	Portugal, Madeira. Wasteland next to an old wall between Madaleno do Mar and Ponta do Sol. NCCPG *Artemisia*. Bernard Tickner, 1994.	NCCPG 1994/90
A. canariensis	Spain, Catalonia, Barcelona, Montjuïc. Botanic Garden of Barcelona. T. Garnatje, M. Veny & S. Garcia, III-2005.	BCN 30424
A. gorgonum	Portugal. Cape Verde Isles, São Antão, upper part of Ribeira da Torre at 1400m (seed via Bonn Botanic Garden, NCCPG Artemisia). W. Lubin, 13-IX-1994.	NCCPG 1995/09
A. vallesiaca	England, United Kingdom. NCCPG *Artemisia*. J. Twibell, 27-06-2005.	NCCPG 1988/30

[a]Vouchers deposited in the herbarium of the Centre de Documentació de Biodiversitat Vegetal de la Universitat de Barcelona (BCN) and in the NCCPG *Artemisia* Collection, Elsworth.

Table 2. Nuclear DNA content and other characters of the populations studied.

Taxa	2C ± s.d. (pg)[a]	2C (Mbp)[b]	Insularity[c]	Domestication[d]
Populations of *Artemisia arborescens*				
Algeria- Gouraya	11.46 ± 0.09	11207.88	Cont.	Wild
Algeria-Essais Hamma	11.32 ± 0.08	11070.96	Cont.	Wild
France-Corsica	11.37 ± 0.12	11119.86	Ins.	Wild
France-Paris	10.97 ± 0.15	10728.66	Cont.	Cult.
France-Porquerolles	11.17 ± 0.13	10924.26	Ins.	Wild
France-Portús	10.37 ± 0.17	10141.86	Cont.	Cult.
Greece-Corfú	10.67 ± 0.08	10435.26	Ins.	Wild
Greece-Crete	11.43 ± 0.11	11178.54	Cont.	Wild
Greece-Rhodes	11.22 ± 0.11	10973.16	Ins.	Wild
Italy-Sardinia	11.30 ± 0.19	11051.4	Ins.	Wild
Italy-Sicily	11.41 ± 0.07	11158.96	Ins.	Wild
Morocco-Rif	11.07 ± 0.07	10826.46	Cont.	Cult.
Spain-Alacant-La Encina	11.22 ± 0.17	10973.16	Cont.	Wild
Spain-Balearic Islands-Formentera (1)	11.20 ± 0.05	10953.60	Cont.	Wild
Spain-Balearic Islands-Formentera (2)	11.28 ± 0.15	11031.84	Cont.	Wild
Spain-Balearic Islands-Mallorca	11.61 ± 0.15	11354.58	Ins.	Wild
Spain-Balearic Islands-Menorca-Binibeca	11.15 ± 0.06	10904.70	Cont.	Wild
Spain-Balearic Islands-Menorca-Maó	11.61 ± 0.23	11354.58	Ins.	Wild
Spain-Barcelona-Nursery "Tres Pins"	10.74 ± 0.24	10503.72	Cont.	Cult.
Spain-Barcelona-Historical Botanic Garden	10.85 ± 0.15	10611.30	Cont.	Cult.
Spain-Botanic Garden of Madrid	10.80 ± 0.13	10562.40	Cont.	Cult.
Spain-Catalonia-Besalú	10.35 ± 0.05	10122.3	Cont.	Cult.
Spain-Catalonia-Roquetes	11.23 ± 0.10	10982.94	Cont.	Wild
Spain-Valencia- Nursery "Pro Agri"	11.15 ± 0.23	10904.7	Cont.	Cult.
Turkey-Samandag	11.18 ± 0.11	10934.04	Cont.	Wild

Cultivars				
"Brass Band"	10.14 ± 0.20	9916.92	-	CV
"Faith Raven" type 1 (from J. Goulsbra, UK)	10.14 ± 0.20	9916.92	-	CV
"Faith Raven" type 1 (from J. Twibell, UK)	10.19 ± 0.07	9965.82	-	CV
"Faith Raven" type 2 (UK)	11.11 ± 0.10	10855.80	-	CV
"Huntington"	9.86 ± 0.07	9643.08	-	CV
"Little Mice" (UK)	9.73 ± 0.05	9515.94	-	CV
"Little Mice" (France-Theix)	9.74 ± 0.20	9525.72	-	CV
"Porquerolles"	11.11 ± 0.14	10865.58	-	CV
"Powis Castle" (UK)	10.29 ± 0.04	10063.62	-	CV
"Powis Castle" (Spain-Barcelona)	10.36 ± 0.10	10132.08	-	CV
Other related species				
A. absinthium (Spain-Catalonia)	8.94 ± 0.04	8743.32	Cont.	Cult.
A. absinthium (Armenia)	8.39 ± 0.40	8205.42	Cont.	Wild
A. absinthium (Iran-Teheran)	8.29 ± 0.10	8107.62	Cont.	Wild
A. absinthium (Spain-Sierra Nevada)	8.79 ± 0.13	8596.62	Cont.	Wild
A. absinthium (France-Mèze)	8.79 ± 0.28	8596.62	Cont.	Cult.
A. argentea	10.30 ± 0.05	10073.40	Ins.	Wild
A. canariensis	10.63 ± 0.17	10396.14	Ins.	Wild
A. gorgonum	10.24 ± 0.17	10014.72	Ins.	Wild
A. vallesiaca	9.81 ± 0.13	9594.18	Cont.	Cult.

[a]2C nuclear DNA content (mean value ± standard deviation of 10 samples). [b]1 pg = 978 Mbp, [c] Cont.= continental populations; Isl.= island populations, [d]Cult.= populations cultivated in gardens, streets, roads or nurseries; CV= cultivars; Wild= wild populations.