



Integration of morphology, genetics, historical and ethnobotanical data: a case of an enigmatic *Genista* (Fabaceae) from Ischia Island (southern Italy)

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The vegetation and flora of Ischia Island (southern Italy) has fascinated several botanists over time, and its peculiar flora, especially on account of the volcanic environments, was carefully studied (e. g. by Gussone 1855, Ricciardi *et al.* 2004). Among the lesser known plants of the island, a species of *Genista* Linnaeus (1753: 709) sect. *Ephedrospartum* Spach (1844: 243) (Fabaceae) occurs in the locality of S. Montano (Lacco Ameno), in the north-western part of the island. It was first identified by Anzalone (1967: 694) as *G. ephedroides* De Candolle (1825: 210), while Ricciardi *et al.* (2004) reported it as *G. gasparrinii* (Gussone 1825: 11) Presl (1826: 19). According to Valsecchi (1993), *G. gasparrinii* is endemic to Sicily, growing in a restricted area of the Gallo Mountain, near Sferracavallo (Palermo province). Ricciardi *et al.* (2004) stated a possible introduction of *G. gasparrinii* from Sicily by G. Gussone, but no documentation for this hypothesis was provided. Brullo *et al.* (1992) indicated *G. gasparrinii* in the Gallo Mountain and in the Tyrrhenian Islands [Aeolian Islands and even Pontine Islands (Latium region, central Italy)]. The populations of these latter archipelagos were previously described as *G. tyrrhena* Valsecchi (1986: 145). On the basis of a specimen collected by B. Anzalone in S. Montano and published by Valsecchi (1993) as *G. tyrrhena*, Conti *et al.* (2007: 44) reported *G. gasparrinii* as endemic to Sicily and as an introduced species in Campania. Recently, Bacchetta *et al.* (2011) proposed two subspecies of *G. tyrrhena*: subsp. *tyrrhena* (endemic to Eolian Islands) and subsp. *pontiana* Brullo & De Marco in Bacchetta *et al.* (2011: 16), endemic to Pontine Islands. The literature data show that the occurrence of *G. gasparrinii* in Ischia Island needs a clarification.

The aim of the present study is to clarify the identity of the Ischia population of *Genista* and its biogeographic relationships with those of *G. gasparrinii* from Sicily (*locus classicus*), using both morphological and molecular approach, coupled with floristic and historical investigations.

Specimen in NAP and photos from CAT and FI (abbreviations according to Thiers 2011) were examined. Field surveys in S. Montano (11 individuals collected) were carried out (see Appendix 1). As regards molecular analyses, the DNA regions chosen refer to one chloroplast DNA intron [*trnL*^(UAA)] and the nuclear genome internal transcribed spacers (ITS1 and ITS2). These markers have been widely used in molecular systematic investigations in species belonging to *G. ephedroides* aggr. (e.g., De Castro & De Luca 2001, De Castro *et al.* 2002, Bacchetta *et al.* 2012). All specimens collected were analysed (see Appendix 1). Total genomic DNA was isolated from approximately 100 mg of fresh leaves following a modified CTAB procedure (De Castro *et al.* 2002). Molecular markers were amplified using primers (Macrogen Inc.) reported in the literature (De Castro *et al.* 2002). The amplified products were purified using a DNA Enzyme-free isolation Spin-Kit (AppliChem GmbH) and purified templates (5 ng) were sequenced according to Di Maio and De Castro (2013), using fluorescent dye (Big Dye™ Terminator Cycle Sequencing Kit ver. 3.1, Applied Biosystems, Life Technologies) and a 3130 Genetic Analyzer (Applied Biosystems, Life Technologies). Complete sequences of both strands of each PCR product were obtained, aligned, and visually checked with the AB DNA Sequencing Analysis ver. 5.2 software (Applied Biosystems, Life Technologies), Sequence Navigator ver. 1.0.1 software (ABI Prism, Perkin Elmer) and BioEdit ver. 7.0.9.0 software (Hall, 1999).