



Primula spectabilis Tratt. aerial parts: Morphology, volatile compounds and flavonoids

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ABSTRACT

The vacuolar and epicuticular flavonoids and the volatiles of the leaves and parts of flower of *P. spectabilis* Tratt., an endemic species in the Italian Oriental Alps, were investigated. From a MeOH extract of the leaves two flavone glycosides, 8-C- β -glucopyranosylluteolin 7-O- α -arabinofuranoside (**1**) and 6-C- α -arabinofuranosylapigenin (**2**) were isolated, in addition to a flavone and three flavonols already known from species of *Primula*. From an EtOH extract of leaf exudates, 7,3',4'-tri-O-methylquercetin was obtained. The structures were elucidated on the basis of their 1D ¹H- and ¹³C NMR data and 2D NMR techniques, as well as of HPLC–MS. The volatiles emitted by the leaves were mainly constituted by non-terpene derivatives, followed by comparable proportions of hemiterpens, oxygenated monoterpenes and sesquiterpene hydrocarbons. In flowers, monoterpene hydrocarbons were the most represented chemical class followed by non-terpene derivatives. Different proportions of compounds were found when individual parts of flowers were examined separately; calyx produced a greater proportion (approx. 49.5%) of non-terpenes as its volatile metabolites.

P. spectabilis has glandular trichomes in the hyaline margins of the epidermal depressions, distributed on the adaxial leaf blade. Glandular hairs were also present on the corolla. Correlations of phytochemical data with the morphological features of leaf, flower and glandular hair are discussed, and a hypothesis is proposed on the ecological roles of the flavonoids and volatile compounds on the general fitness of the species and cross-pollination strategies.

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1. Introduction

Primula L. genus (Primulaceae) comprises 426 species distributed in temperate and cold regions of the Northern hemisphere and in mountains at tropical latitudes.

Primula spectabilis Tratt. is an endemic species of the Eastern Alps (from Monte Grappa to the Brescia Pre-Alps), protected by the L.R. 27/09/77 n. 33 for the spontaneous flora. It only lives between 600 and 2500 m above the sea level, on humid and shaded rocks or stony and pebbly slopes (Ravazzi, 1999; Richards, 2003). This species belongs to sect. *Auricula* Duby and is characterized by involute leaf veneration and stomata on the adaxial surface of the leaf, as reported by Smith and Forrest (1929).

The leaves are coriaceous, rhombic-ovate and pressed to the ground, and display glandular hairs on the upper surface, embedded in the parenchyma and in the cartilaginous margin. The scape

is 2–15 cm in height, bearing 2–5 pinkish-red to lilac flowers with pedicels of 1–2 cm and a calyx of 7–12 mm; the bracts are triangular-lanceolate to linear (Pignatti, 1982; Tutin et al., 1993). The glands of *Primula* species produce farinas (formed as rod or needle-like crystals) and/or exudates (sticky secretions); in particular, the leaves of *P. spectabilis* produce an exudate deposition. Although the morphology of glandular hairs of some Italian *Primula* species has already been reported (Higuchi et al., 1999; Fico et al., 2007), no specific study on *P. spectabilis* has been published so far.

The secondary metabolite content of *Primula* genus has not been completely characterized yet. Earlier studies reported the presence of saponins, especially in rhizome and roots (Calis, 1992; Morozowska and Wesoowska, 2004), responsible for its medicinal use as expectorant and antimicrobial drug (Della Loggia, 1993). Many works deal with the flavonoid composition of some *Primula* species: *P. vulgaris* (Harborne, 1968), *P. elatior* (Petitjean-Freytet, 1993), *P. veris* (Huck et al., 2000), *P. denticulata* (Tokalov et al., 2004), *P. hirsuta*, *P. auricula* and *P. daonensis* (Fico et al., 2007). Harborne (1968) used data on flavonoids in order to determine the relationship between flavonoid content and taxonomy in the *Primulaceae*. Among flavonols, mono- and diglycosylated

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derivatives characterize this genus. The most common sugars are glucose, rhamnose, xylose and galactose, while the main aglycons are quercetin, kaempferol and isorhamnetin; on average, sugar moiety linkage favours the 3-position.

This is the first study on the hair morphology of the Italian endemic species *P. spectabilis* and its vacuolar and epicuticular flavonoids. Moreover, the volatile compounds emitted by the leaves, the whole flower and its parts are also investigated.

2. Results

2.1. Aerial part structure, trichome types, morphology and distribution

The leaves of *P. spectabilis* show a rhombic-ovate outline and are coriaceous and thickly imbricate, forming a basal rosette (Fig. 1). They are distinguished by a white (hyaline), cartilaginous margin which folds up to the upper epidermis. The adaxial lamina is densely covered with glandular trichomes producing sticky secretions with a strong aromatic smell. They are embedded and randomly interposed among stomata (Fig. 2A and B). Hairs can also be found on the abaxial surface and along the margins.



Fig. 1. *P. spectabilis* whole plant.

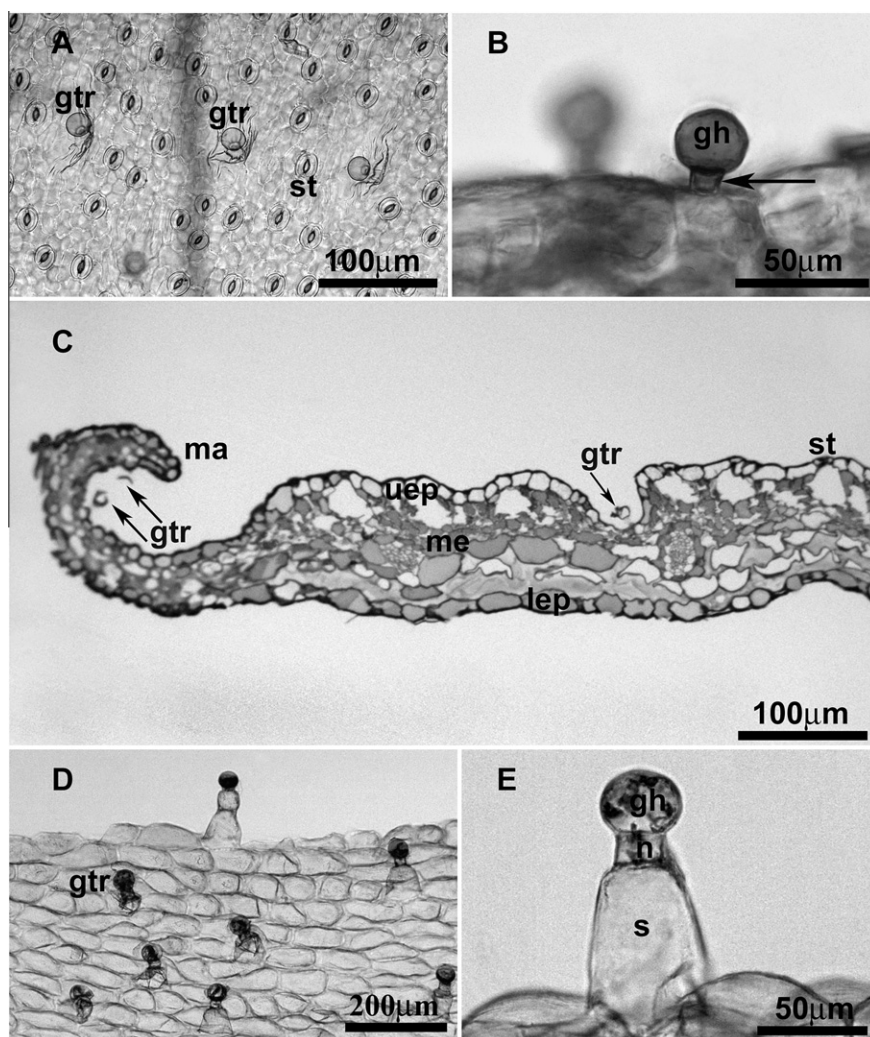


Fig. 2. LM micrograph of epistomatic leaf of *P. spectabilis*: both stomata and glandular trichomes are in the upper epidermis (A); LM micrograph of a single big-headed leaf glandular trichome (B); LM micrograph of transverse section of an epistomatic leaf of *P. spectabilis*: the glandular trichomes are located in the epidermal crypts; numerous glandular hairs can also be found on the involute leaf margin (C); LM micrograph of glandular hairs of *P. spectabilis* petals with long conical stalks and round glands (D, E). Abbreviations: gtr, glandular trichome; gh, glandular head; st, stomata; uep, upper epidermis; lep, lower epidermis; ma, margin; me, mesophyll; n, neck; s, stalk.

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