A new orchid hybrid *Dactylorhiza* ×silvae-gabretae (D. fuchsii × D. traunsteineri) from the Czech Republic

Nový kříženec Dactylorhiza ×silvae-gabretae (D. fuchsii × D. traunsteineri) z České republiky

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Procházka F. & Čurn V. (1998): A new orchid hybrid *Dactylorhiza* × silvae-gabretae (D. fuchsii × D. traunsteineri) from the Czech Republic, – Preslia, Praha, 70: 235–240.

An orchid population of *Dactylorhiza traunsteineri* in the southeastern part of the Czech Bohemian Forest (Šumava Mts) in southwestern Bohemia (near the village Želnava) was studied for more than 20 years. Besides the hybrid of this species with *D. comosa* subsp. *majalis* (= *D. ×dufftiana*), plants corresponding with the hybrid combination *D. fuchsii × D. traunsteineri* were observed. This new hybrid is described as *Dactylorhiza ×silvae-gabretae* Procházka et Čurn. Isozyme markers separated by nondenaturing discontinuous polyacrylamide gel electrophoresis were used for description and classification of plants belonging to *D. traunsteineri*, *D. fuchsii* and *D. ×silvae-gabretae*. The pattern of SDH (shikimate dehydrogenase) isozymes exhibited a hybrid character in analysed plants of *D. ×silvae-gabretae*.

K e y w o r d s: Nomenclature, taxonomy, *Dactylorhiza*, hybridization, isozyme analyses, Bohemian Forest, Southwest Bohemia, Czech Republic

The extremely rare Central European orchid *Dactylorhiza traunsteineri* (Sauter) Soó was first found in the Šumava Mts in two localities by S. Kučera in the second half of the 1970s. M. Fassati found this orchid approximately 15 years earlier in an unspecified locality in the southeastern part of the Šumava Mts (J. Holub, personal communication). Both localities (near Želnava and Záhvozdí) are located in the Czech part of the mountains, approximately 1.5 km from each other. More than 10 years later (in 1989), the species was also found in Bavaria, in the peat-bog Klosterfilz-St. Oswald-Riedlhütte (Procházka in Kováříková 1998). All three localities (two located in the quadrant 7149d and one in the quadrant 7046c of the phytogeographical mapping grid) are at 735–740 m a. s. l. The distribution in the Bohemian, Austrian and Bavarian parts of the Šumava Mts is shown in Fig. 1.

The locality of *D. traunsteineri* near Želnava was regularly monitored since the second half of the seventies by the senior author (Procházka 1980, 1983) who found there the primary hybrid *D.* × *dufftiana* (Schulze) Soó (= *D. comosa* subsp. *majalis* × *D. traunsteineri*) in 1980 (together with M. Hájek, Choceň). For several years, plants of *Dactylorhiza fuchsii* (Druce) Soó were also observed there, and plants corresponding in their morphology to the hybrid *D. fuchsii* × *D. traunsteineri* were rarely being found as well (cf. Procházka 1983: 242).

In 1994, two individuals belonging to this hybrid were found. During excursions in 1995–1997, the plants were studied also by V. Faltys, J. Kováříková, A. Pavlíčko and M. Štech. Because this hybrid has not been validly described yet, its description follows:

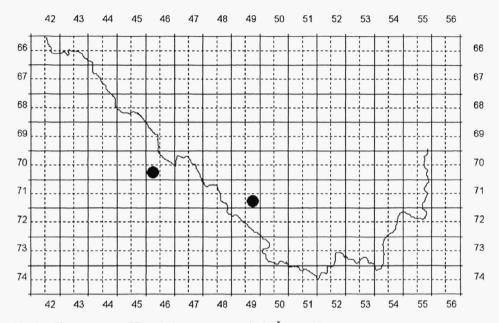


Fig. 1. - The occurrence of Dactylorhiza traunsteineri in the Šumava Mts.

Dactylorhiza × silvae-gabretae Procházka et Čurn, hybr. nova

 $(= Dactylorhiza fuchsii subsp. fuchsii \times D. traunsteineri)$

Diagnosis: Planta habitu media inter parentes (*Dactylorhiza fuchsii* subsp. *fuchsii* et *D. traunsteineri*), 35–60 cm alta; tuberibus (2) 3–4-fidis, apicibus breviter radiciformiter attenuatis vel valde elongatis; caule stricto, anguste cavo, laxe foliato, in basi 5–7 mm in diametro; foliis (plerumque praeter tria basalia squamoidea sine colore viridi) tribus usque ad quatuor solum (super illis maiori parte etiam 2–3 (4) folia parva bracteis similia sine basi vaginata), erecto-patentibus, suberectis, lanceolato-linearibus, acutis, subplicate carinatis, maculatis, iis medianis (12) 13–16 (18) cm longis, in medio fere 14–22 mm latis; racemo erecto, breviter cylindraceo, mediocriter dense multifloro, 7–14 cm longo; floribus mediocriter magnis, lucide purpureis. Labellum semper latius quam longum, trilobum, atropurpuree pictum. Bracteae plerumque evidenter longitudinem florum excedentes. Pollinia plerumque obscure rubra.

Florendi tempus: pars secunda mensis Iunii usque ad Iulium.

I conotypus: Bohemia meridionalis, montes Silvae Gabretae (Šumava, Czech Bohemian Forest, Böhmerwald): campus fluvii Vltavae ca 1,2 km ad septentrio-occidentem a pago Želnava versus, 735 m s. m., rare cum parentibus, 30. VI. 1995, V. Čurn; phototypus hoc loco in plate Ib et IIb (additamentum color.).

Several authors already reported the hybrid combination *D. fuchsii* × *D. traunsteineri*. This combination was even named twice, but invalidly (*D.* ×*kelleriana* P. Hunt, nom. inval., Orchid Reviews 79: 139, 1971; *D.* × *robertsii* Averyanov, nom. inval., Bot. Zhurn. 71: 93, 1986).

The hybrid *Dactylorhiza* × *silvae-gabretae* (Fig. 2) has been recorded in northwestern Wales, Yorkshire and Ireland. However, plants from these regions represent another morphological type: "This hybrid has the dwarf habit and narrow leaves of *D. traunsteineri*, which are often marked with dark purple, narrow transverse bars or transversely elongated spots or rings. The flowers have a trilobed labellum which is nearer *D. fuchsii* in its markings. The pollen is highly sterile" (Sell & Murrell 1996: 346).





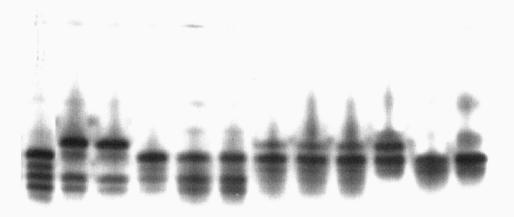








Fig. 2. – Parental species and their hybrid: the plants and inflorescence details. Previous page: *Dactylorhiza fuchsii* (top), *D. silvae-gabretae* (bottom). This page: *D. traunsteineri*.



1 2 3 4 5 6 7 8 9 10 11 12

Fig. 3. – Pattern of SDH isozymes in analysed samples. 1. *Dactylorhiza majalis*, Horská Kvilda; 2. – 3. *D. trausteineri*, Želnava; 4. – 6. *D. traunsteineri* (×*D. majalis*), Želnava; 7. – 9. *D. ×silvae-gabretae*, Želnava; 10. – 12. *D. fuchsii*, Želnava.

According to our opinion, based on 20 years of observations, a very strong introgression with *D. comosa* subsp. *majalis* (Reichenb.) P. D. Sell occurs in the locality, despite the fact that *D. comosa* subsp. *majalis* is not present there in a genetically pure form (it occurs several hundred metres apart). In the late seventies, plants morphologically corresponding to *D. traunsteineri* prevailed (particularly in a gap in alder carr). These morphotypes are rare now, and the population contains mainly plants exhibiting also characters of *D. comosa* subsp. *majalis*.

Biochemical markers (isozymes or proteins without detectable enzymatic activity), separated after electrophoresis into discrete bands, are useful tools for characterization of biochemical variability in a population. These markers also have application in the field of identification and description of genotypes, in population genetics, and in taxonomic and phylogenetic studies (Tanksley & Orton 1983, Soltis & Soltis 1990).

Plants belonging to three taxa were used for analyses of biochemical markers: morphologically typical plants of D. traunsteineri, two hybrid plants D. × silvae-gabretae, and plants of D. fuchsii occurring at the same locality. Enzymes were extracted from fresh green leaves (2 % glutathione extraction buffer), separated using non-denaturing discontinuous PAGE system (4 % stacking gel, pH = 6.8, 7.5 % separating gel, pH = 8.8, Tris-glycine electrode buffer, pH = 8.3), and enzymatic activity on gel was detected. Method of digital image analysis was used to evaluate zymograms. Methods of biochemical and digital analysis are described in Čurn (1995) and Čurn & Sáková (1996).

Results of SDH isoenzyme analyses showed that *D. traunsteineri* (the morphologically typical plants) contain only alleles 'b' and 'd' in its genotype. *D. traunsteineri* is also characterized by the absence of 'a' and 'c' alleles (allele 'a' should be found in *D. comosa* subsp. *majalis*). Higher genetic variation was found in analysed plants of *D. fuchsii*. Analysed plants exhibited presence of the 'c' allele only, or in some plants, of the 'c' and 'd' alleles. Hybrid taxon *D. × silvae-gabretae* was characterised by the presence of 'c' allele, originated from *D. fuchsii*. One of the analysed plants exhibited allelic constitution 'ccdd'; the second plant has present also 'b' allele and allelic constitution 'ddcb' (Fig. 3). These results conform the hybrid origin of plants reported in the present study.

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Souhrn

Déle než 15 let byly na známé šumavské lokalitě *Dactylorhiza traunsteineri* u Želnavy objevené v 70. letech S. Kučerou, ojediněle pozorovány rostliny, které se zdály být křížencem mezi *D. fuchsii* a *D. traunsteineri*. Na základě rostlin objevených na lokalitě v roce 1995 byl popsán nový kříženec *Dactylorhiza* × *silvae-gabretae* Procházka et Čurn. Objektivní existence tohoto křížence byla prokázána isoenzymovou analýzou.

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