

AN OVERVIEW OF RECENTLY DISCOVERED INTRA AND INTER-GENERIC ORCHID HYBRIDS AS NEW ADDITIONS TO ROMANIAN FLORA (Part II)

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Abstract

Nine orchid hybrids, previously unrecorded in Romanian flora, have been identified. These hybrids include both intra-generic (within the same genus) and inter-generic (between different genera) crosses, showing the extraordinary biodiversity within the region. Such hybrids are extremely rare, not only in Romania but throughout temperate Europe, making this discovery particularly significant. During our eight-year study (2017-2024), we encountered several hybrids belonging to various orchid genera (*Anacamptis*, *Ophrys*, *Gymnadenia*, *Dactylorhiza*, *Epipactis*) and nothogenera (\times *Dactylocamptis* and \times *Dactylanthera*). Of these nine orchid taxa, eight are regarded as new to science, being documented for the first time. These findings are part of an ongoing research project – Orchids of Romania Project, which continues to gather additional data on these unique hybrids. The study includes detailed information on the hybrids' distribution, habitat, ecology, phenology and conservation status as assessed by the IUCN.

Key words: orchids, hybrids, conservation, intrageneric, Romania.

INTRODUCTION

Over millions of years of evolution, the process of hybridization has significantly impacted the history of life on Earth (Angheliescu et al., 2020). In the natural environment, hybridization serves as an essential evolutionary mechanism that, in the case of the Orchidaceae family, transcends reproductive barriers between different species and even between different genera (Angheliescu et al., 2020). Following cross-pollination, new hybrid lines are formed through the recombination of parental genomes (Angheliescu et al., 2021a). The concept of nothotaxon refers to the totality of hybrids resulting from the crossing of two distinct parental taxa (natural species, excluding hybrids). These nothotaxa can subsequently evolve into higher taxonomic entities such as nothogenus, nothospecies, or nothosubspecies (Angheliescu et al., 2021a). Thus, hybridization represents a crucial element in the global speciation process, significantly contributing to the formation of new species with enhanced genetic and adaptive variation (Kretzschmar et al., 2007). The Orchidaceae

family is unique in producing natural hybrids between species belonging to the same genus (intrageneric hybrids), as well as hybrids resulting from the crossing of two or more species from different genera (intergeneric hybrids) (Angheliescu et al., 2023a). Hybridogenic populations, also known as nothopopulations, can be very numerous and widespread over large areas, sometimes competing with parental populations. In this article, we document the first reported natural occurrence of seven intrageneric hybrids belonging to several orchid genera: *Anacamptis* - *Anacamptis* \times *menosii* nothosubsp. *angelliana* N. Angheliescu, N. Kigyosy, L. Balogh & Mih. Balogh, 2024 nothosubsp. *nov.* (*Anacamptis coriophora* subsp. *coriophora* \times *Anacamptis papilionacea*), *Anacamptis* \times *nicodemi* nothosubsp. *caucasica* N. Angheliescu, L. Balogh & Mih. Balogh, N. Kigyosy, 2024 nothosubsp. *nov.* (*Anacamptis morio* subsp. *caucasica* \times *Anacamptis papilionacea*), *Dactylorhiza* - *Dactylorhiza* \times *subalpina* N. Angheliescu, L. Balogh & Mih. Balogh, N. Kigyosy, 2024 nothosp. *nov.* (*Dactylorhiza*

cordigera ssp. *sicolorum* × *Dactylorhiza incarnata*), *Dactylorhiza* × *carpathiana* (Soó) Soó N. Anghelescu, L. Balogh & Mih. Balogh, N. Kigyosy, 2024 nothosp. nov. (*Dactylorhiza cordigera* × *Dactylorhiza majalis*) and *Epipactis* - *Epipactis* × *romanensis* R. Maalouf, N. Anghelescu, L. Balogh & Mih. Balogh, N. Kigyosy, 2024 nothosp. nov. (*Epipactis distans* × *Epipactis atrorubens*), *Gymnadenia* - *Gymnadenia* × *montana* N. Anghelescu, L. Balogh, M. Balogh & N. Kigyosy, 2024 nothosp. nov. (*Gymnadenia winkeliana* × *Nigritella nigra* subsp. *bucegiana*) and *Ophrys* - *Ophrys* × *minuticauda* nothosubsp. *cornuta* R. Maalouf, N. Anghelescu, L. Balogh & Mih. Balogh, N. Kigyosy, 2024 nothosubsp. nov. (*Ophrys apifera* × *Ophrys scolopax* subsp. *cornuta*). Additionally, we included two rare intergeneric hybrids, members of the nothogenera × *Dactylocamptis* - × *Dactylocamptis* *suzana* N. Anghelescu, L. Balogh & Mih. Balogh, N. Kigyosy, 2024 nothosp. nov. (*Anacamptis coriophora* × *Dactylorhiza maculata*) and × *Dactylanthera* × *Dactylanthera transsilvanica* L. Balogh & Mih. Balogh, N. Anghelescu, N. Kigyosy, 2024 nothosp. nov. (*Dactylorhiza maculata* × *Platanthera bifolia*).

The likelihood of these specific hybridization events occurring in the wild and successfully producing viable individuals is exceedingly rare. This rarity highlights the exceptional nature of their discovery and underscores the unique conditions that must align for such hybrids to form in the wild. Out of these nine newly described hybrids, eight are regarded as new-to-science taxa, hereby described for the first time. The intrageneric hybrid *Epipactis* × *romanensis* R. Maalouf, N. Anghelescu, L. Balogh & Mih. Balogh, N. Kigyosy, 2024 nothosp. nov. (*Epipactis distans* × *Epipactis atrorubens*) featured under the name *Epipactis* × *gauckleri* in two previous publications (Weigel & Riechelmann, 2002), as mentioned in Günther Blaich's List of Hybrids of European Orchids (2024). However, the taxon *Epipactis* × *gauckleri* does not feature as an accepted taxon on Kew's database, Plants of the World Online (POWO, accessed 2024), the database which we take as a reference for all our taxonomical studies. Consequently, we chose to make a formal description of this taxon with the

confidence that it will be listed shortly on the international plant databases (POWO, WFO, 2024). At the same time, two Romanian endemic orchid taxa (intrageneric hybrids), *Anacamptis* × *menosii* nothosubsp. *angelliana* N. Anghelescu, N. Kigyosy, L. Balogh & Mih. Balogh, 2024 nothosubsp. nov. (*Anacamptis coriophora* subsp. *coriophora* × *Anacamptis papilionacea*) and *Ophrys* × *minuticauda* nothosubsp. *cornuta* R. Maalouf, N. Anghelescu, L. Balogh & Mih. Balogh, N. Kigyosy, 2024 nothosubsp. nov. (*Ophrys apifera* × *Ophrys scolopax* subsp. *cornuta*), described previously by Anghelescu et al. (2023a), are reconsidered in this paper and redescribed as new to science taxa, following the kind and helpful advice of IPNI editors at Royal Botanic Gardens, Kew.

During our eight-year study from 2017 to 2024, we carried out extensive fieldwork across various protected areas, including national and natural parks, to document and map the orchid flora found in the newly identified remote hybridogenic zones (Anghelescu et al., 2020; 2023a; 2023b; De Anghelli & Anghelescu 2020, 2023).

Studying orchid hybrids presents numerous challenges due to their complex genetic compositions and varied phenotypic expressions. Many hybrids exhibit a wide range of morphological variations, making it difficult to establish clear identification criteria (Anghelescu et al., 2020). Additionally, the often-limited availability of individuals for study, particularly in natural habitats where rare orchid hybrids are found, further complicates research efforts. Environmental factors, such as habitat conditions and pollinator interactions, also play a significant role in the success of hybridization, adding layers of complexity to the investigation. Furthermore, the legal protections surrounding many orchid taxa restrict access to specimens, which can hinder comprehensive research and the development of a complete understanding of hybrid dynamics. These obstacles necessitate a meticulous and adaptable approach to studying orchid hybrids, requiring both patience and innovative methodologies. It is important to highlight that collecting rare plant taxa, including our rare orchid hybrids from strictly protected areas and national parks, is illegal, even if a study or work permit is

obtained. These taxa are safeguarded by both international laws (CITES) and regional Romanian regulations. Additionally, for laboratory research or herbarium voucher deposits, the collection is limited to only 1-3 individual flowers and, in exceptional cases, a single leaf from the holotype. In recent years, photos of the holotype(s) have also been accepted as herbarium vouchers to further ensure the protection and preservation of these extremely rare and vulnerable specimens (Anghelescu et al., 2024).

The objectives of the current study are as follows: (1) to provide a detailed description of the primary morphological traits of the two nothospecies, particularly their floral characteristics, in comparison to their parent species; (2) to offer insights into various biological aspects that are crucial for understanding their ecological needs, including distribution, habitat, community interactions, phenology, reproductive requirements, and conservation efforts; and (3) to report on the IUCN (2021) conservation status of all the considered taxa, accompanied by photographs of the holotypes. Given the significance and rarity of these newly identified nothotaxa, we strongly advocate for the inclusion of these two nothopopulations as new additions to the Romanian flora.

MATERIALS AND METHODS

Sites Studied

The hybrids were found/studied in two major protected natural areas of Romania, Bucegi Natural Park ROSCI0013, Prahova County (Hedrén et al., 2022; Anghelescu et al., 2023b) and Harghita Mădăraș, ROSCI00090, Harghita County, as well as in various other regions known for their rich orchid diversity, such as Prahova, Harghita and Mehedinți Counties:

(1) Bucegi Natural Park, Southern Carpathians, Central Romania, is a protected area included within Natura 2000 ROSCI0013, IUCN category V. It covers Prahova, Dâmbovița and Brașov Counties, with an area of ca. 32,663 ha/326.63 km² and the highest elevation at Omu Peak of 2,505-2,514 meters a.s.l (Ielencz, 2005; Herdren et al., 2022).

(2) Natura 2000 protected area ROSCI00090 Harghita-Mădăraș, located in the central-eastern

part of Harghita County (EU Environment, 2014) and covers an area of 13,373 hectares. It is located at an altitude between 1,500-1,800 meters a.s.l (above sea level) (Mikfalvi & Vifkori, 1979; Marcu, 1986). The site is a natural area covered by deciduous, coniferous and mixed forests, natural meadows, heathlands, bogs (peat bogs) and steppes together with a rich hydrographic network that consists of several lakes and watercourses (Ciocârlan 2000; 2009).

(3) Mehedinți County - the species studied prefer full sun, grassy, alkaline marsh, prone to flooding (wet meadows), forest clearings, on calcareous substrates, up to 100 meters a.s.l.

Morphometric methods/comparisons

Particular emphasis was placed on taxonomically informative characteristics, especially those that differentiate the morphology of the leaves, labellum, and tepals. Measurements of both vegetative and floral parts were taken from living plants and fresh flowers (Anghelescu et al., 2021a; 2023a). Our observations indicate that the primary floral traits of the nothotaxa are generally intermediate between those of the parent species.

Digital Photographic Equipment

Digital images of individual plants and floral parts were taken using Canon 5D Mark III, Nikon D3 and Nikon D850 camera bodies equipped with Nikon Micro NIKKOR 60 mm and NIKKOR 24.0-70.0 mm lenses, Venus Optics Laowa 100 mm 2X Ultra Macro. Additional equipment included a Manfrotto Tripod and Litra Torches 2.0s. Images were analysed using Adobe Photoshop® CC 2023, Zerene Stacker Software, Vers.2021-11-16 (Anghelescu et al., 2021b, 2023a, 2024).

Taxonomical classification

The taxonomy is structured according to the most recent databases in line with the International Plant Names Index n.d. (IPNI, 2024), Plants of the World Online | Kew Science, n.d. (POWO Kew, 2024), The WFO Plant List | World Flora Online, n.d. (WFO, 2024), New Taxa | Euro+Med-Plantbase, n.d. (2024), De Angelli & Anghelescu (2020, 2023).

RESULTS AND DISCUSSIONS

This overview examines the recent discoveries of intra- and inter-generic orchid hybrids that have been identified as new contributions to the flora of Romania. These hybrids, resulting from the complex interplay of various species within the Orchidaceae family, not only enrich the botanical diversity of the region but also highlight the evolutionary dynamics at play within these fascinating plants. Recent studies have focused on documenting these hybrids and their morphological characteristics, ecological requirements, and distribution patterns (Anghelescu et al., 2020; 2021a; 2021b; 2023a). Intra-generic hybrids have emerged from crossings within the same genus, showcasing unique adaptations and traits that distinguish them from their parent species. On the other hand, inter-generic hybrids, formed by the combination of distinct genera, reveal the remarkable reproductive flexibility and resilience of orchids in natural conditions. As a result, hybridization in natural conditions serves as an evolutionary catalyst, breaking down the reproductive barriers that divide different species. By definition, hybridization involves the mating or cross-breeding of two parental lineages with distinct genetic backgrounds. In the first generation of offspring, known as F1 or primary hybrids, their genes combine to create intermediary forms between the parent taxa (Scopece et al., 2007). This phenomenon can have significant impacts on genetic variation within populations and the evolution of species, leading to hybrids with unique combinations of traits (Marques et al., 2014). In some cases, these hybridization events may even result in the formation of entirely new nothotaxa, such as nothogenera, nothospecies, and nothosubspecies (Mayr, 1942).

Description of the nine nothotaxa studied

Following is the full description of the nine intra- and intergeneric orchid hybrids included in this study, named according to POWO (2024). The included nothotaxa are all terrestrial, perennial, rhizomatous, autotrophic, sympodial herbaceous geophytes (Figures 1-9).

Anacamptis × *menosii* nothosubsp. *angelliana*
N. Anghelescu, N. Kigyosy, L. Balogh & Mih. Balogh, 2024 nothosubsp. nov. (Figure 1).

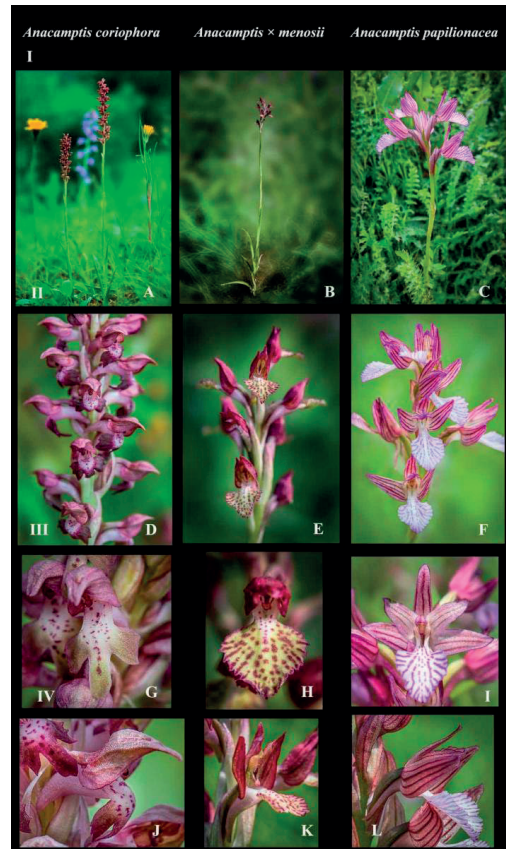


Figure 1. I. Full plants in their natural habitats; II. Inflorescences details; III. Individual flower detail; IV. Side-view of individual flower details. A., D., G. J. - *Anacamptis coriophora*; B., E., H. K. - *Anacamptis* × *menosii* nothosubsp. *angelliana*; C., F., I. L. - *Anacamptis papilionacea*. Photos © Nora E. Anghelescu, 31 May 2023, Mehedinți County

Hybrid formula: *Anacamptis coriophora* subsp. *coriophora* × *Anacamptis papilionacea*.

Discovered: 31 May 2023, Nora E. Anghelescu.

Locus classicus: Mehedinți County, private property.

Flowering time: May-June.

Native to: Romania, Endemic (En).

Description: Stem (epigeal), 10-14 cm, erect, vivid green, with no purple pigmentation. 1-2 basal leaves sheath the stem, 3-5 cauline unspotted leaves are sessile, vivid green, acuminate, sheathing the stem (resembling *Anacamptis coriophora*; in *Anacamptis papilionacea*, the leaves are veined and purpletinged at the tips). The inflorescence is a lax raceme (resembling *Anacamptis papilionacea*), less floriferous, with 8-10 medium-sized flowers

resembling *Anacamptis coriophora*. The helmet is deep-purple pigmented and tight, with an elongated tip, resembling *Anacamptis coriophora*. The labellum is entire and heart-shaped (in upper flowers, resembling *Anacamptis papilionacea*) to mildly three-lobed (in basal flowers, resembling *Anacamptis coriophora*), smaller in size than *Anacamptis papilionacea*. Labellar stripes and dots are deep-purple to red (heterosis effect), resembling *Anacamptis papilionacea*. The spur is conical and thick, resembling *Anacamptis coriophora* (correction of the original taxonomical description in Anghelescu et al., 2023a, pg. 421).

Nothopopulation counts: 1(2) individuals/hybrids, flowering each year.

Herbarium voucher specimen: Deposited at the Herbarium of the Botanical Garden Bucharest, N.E. Anghelescu, barcode: **BUC 410363**.

Proposed conservation status: Endangered (EN).

Anacamptis × nicodemi nothosubsp. *caucasica*
N. Anghelescu, L. Balogh & Mih. Balogh, N. Kigyosy, 2024 nothosubsp. *nov.* (Figure 2)

Hybrid formula: *Anacamptis morio* subsp. *caucasica* × *Anacamptis papilionacea*.

Discovered: 22 May 2019, Nora E. Anghelescu.

Locus classicus: Mehedinți County, private property.

Flowering time: May-June.

Native to: Romania, Endemic (En).

Description: Stem (epigeal), 9-15(25) cm, erect, vivid green, with no purple apical pigmentation. 4-6 basal leaves, of which 2-3 sheath the stem; 2-4 cauline unspotted leaves, sessile, vivid green, acuminate, sheath the stem, resembling *Anacamptis papilionacea*. Inflorescence is a lax raceme, resembling *Anacamptis papilionacea*, less floriferous, with 8(10)-15(20) medium-sized flowers resembling either *Anacamptis papilionacea* and/or *Anacamptis morio* subsp. *caucasica*. No hybrid is as variable in the shape and colour of the flowers as *Anacamptis × nicodemi* nothosubsp. *caucasica*, since it inherits the enormous colour variability (and genetic plasticity) of its parent, *Anacamptis morio* subsp. *caucasica*. Furthermore, the influence of the morio parent is recognized in the slightly ascending spur and the fan-like labellum adorned with various hues of

purple markings. Nevertheless, the radial arrangement of the markings reminds of *Anacamptis papilionacea*. From the latter parent, it also inherited the drop-like shape, the lighter colour and the serrated edges of the labellum. The hood is also tighter, similar to *Anacamptis papilionacea*. The horizontally to upward-oriented spur (a feature inherited from *Anacamptis morio* subsp. *caucasica*) is the main characteristic that differentiates it from *Anacamptis × nicodemi*, which typically has a slightly downward-oriented spur (a feature inherited from *Anacamptis morio* subsp. *morio* parent).

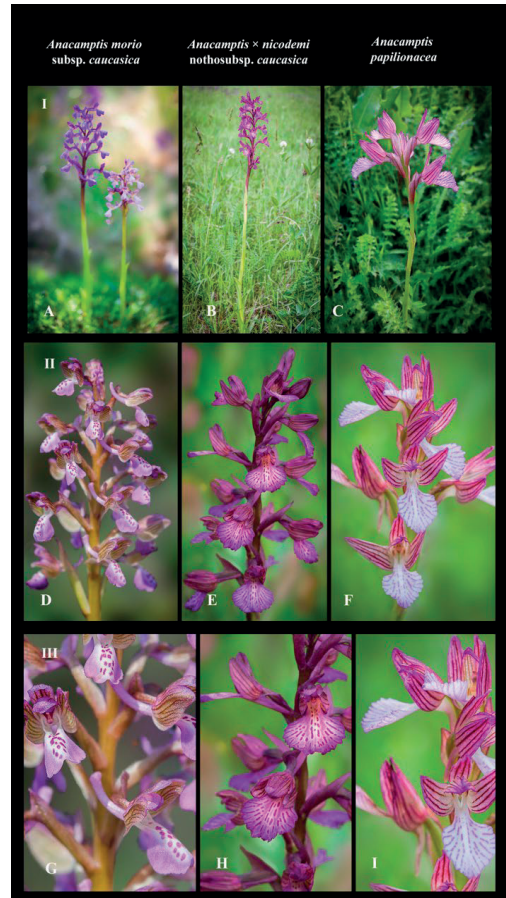


Figure 2. I. Full plants in their natural habitats; II. Inflorescences details; III. Individual flower detail; A., D., G. - *Anacamptis morio* subsp. *caucasica*; B., E., H. - *Anacamptis × nicodemi* nothosubsp. *caucasica*; C., F., I. - *Anacamptis papilionacea*. Photos © Nora E. Anghelescu, 22 May 2019, Mehedinți County

Nothopopulation counts: numerous, >100 individuals/hybrids, flowering each year.

Herbarium voucher specimen: Deposited at the Herbarium of the University of Agronomic Sciences and Veterinary Medicine of Bucharest, N.E. Anghelescu, **USAMVB–barcode 40104**.

Proposed conservation status: Endangered (EN).

Dactylorhiza × *subalpina* N. Anghelescu, L. Balogh & Mih. Balogh, N. Kigyossy, 2024 nothosp. nov. (Figure 3).

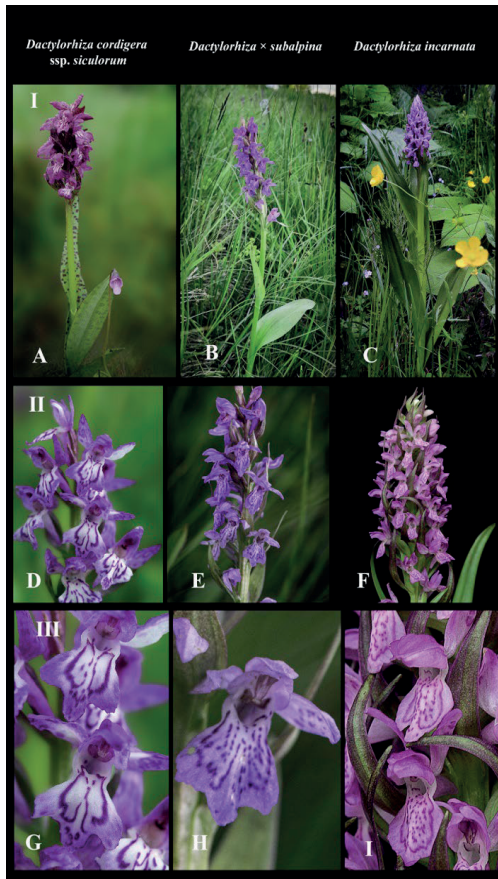


Figure 3. I. Full plants in their natural habitats; II. Inflorescences details; III. Individual flower detail; A., D., G. - *Dactylorhiza cordigera* subsp. *siculorum*; B., E., H. - *Dactylorhiza* × *subalpina*; C., F., I. - *Dactylorhiza incarnata*. Photos © Nora E. Anghelescu, 31 May 2022, Harghita County

Hybrid formula: *Dactylorhiza cordigera* ssp. *siculorum* × *Dactylorhiza incarnata*.

Discovered: 24 June 2023, Nora E. Anghelescu.

Locus classicus: Harghita Mădăraș, ROSCI00090.

Flowering time: June–July.

Native to: Romania, Endemic (En).

Description: The stem (epigeal), 22–36 cm is erect, slender, vivid green, reminiscent of *Dactylorhiza incarnata*, with purple apical pigmentation, resembling *Dactylorhiza cordigera* subsp. *siculorum*. The basal leaf is wide, vivid green and mostly unpigmented, resembling *Dactylorhiza incarnata*. It is broadest in the middle, a feature inherited from *Dactylorhiza cordigera* subsp. *siculorum*. The two sessile, unspotted cauline leaves are vivid green and sheath the stem; the upper one is narrow-acuminate, bract-shaped. The inflorescence is an elongated, lax raceme similar to that of the *Dactylorhiza cordigera* subsp. *siculorum* parent. The flowers are medium-sized, purple-coloured, intermediate between the two parental species. The three-lobed labellum is also intermediate in shape and pigmentation between parents.

Its lateral, scalloped lobes are shorter than the median lobe, spread horizontally, resembling *Dactylorhiza cordigera* subsp. *siculorum*. The labellum is narrower, ovoidal and more elongated than the heart-shaped labellum of *Dactylorhiza cordigera* subsp. *siculorum*, resembling more *Dactylorhiza incarnata* parent. The labellar markings form circular double or triple loops, further resembling *Dactylorhiza incarnata*. The spur is longer than that of *Dactylorhiza cordigera* subsp. *siculorum*, conical, horizontal to slightly downward pointing, resembling *Dactylorhiza incarnata*. Overall, the hybrids phenotypically resemble the *Dactylorhiza incarnata* parent more closely. This nothopopulations is still under study.

Nothopopulation counts: 3 individuals/hybrids, flowering each year.

Herbarium voucher specimen: Deposited at the Herbarium of the University of Agronomic Sciences and Veterinary Medicine of Bucharest, N.E. Anghelescu, **USAMVB–barcode 40105**.

Proposed conservation status: Endangered (EN).

Dactylorhiza × *carpathiana* (Soó) Soó N. Anghelescu, L. Balogh & Mih. Balogh, N. Kigyossy, 2024 nothosp. nov. (Figure 4).

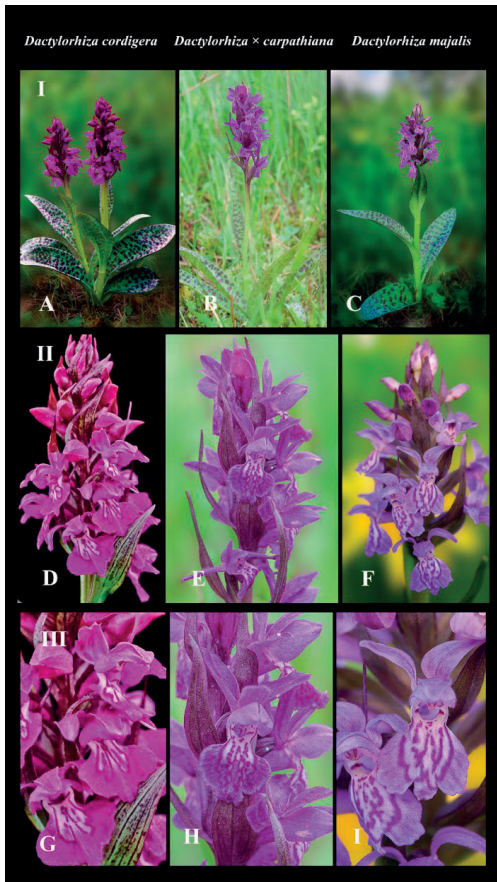


Figure 4. I. Full plants in their natural habitats; II. Inflorescences details; III. Individual flower detail; A., D., G. - *Dactylorhiza cordigera*; B., E., H. - *Dactylorhiza* × *carpathiana*; C., F., I. - *Dactylorhiza majalis*. Photos © Nora E. Anghelescu, 5 June 2018, Prahova County

Hybrid formula: *Dactylorhiza cordigera* × *Dactylorhiza majalis*

Discovered: 5 June 2018, Nora E. Anghelescu.

Locus classicus: Bucegi Natural Park ROSCI0013.

Flowering time: May-June.

Native to: Romania, Endemic (En).

Description: The stem (epigeal), 15-32 cm is erect, slender, deep-green, significantly purple-pigmented in the rachis (inflorescence) part, similar to both parents. The lowest/most basal leaf is vivid green and broadest toward the middle, with a more roundish apex, resembling *Dactylorhiza majalis*. The 2-3 basal leaves are strongly purple pigmented on the upper side, with irregular maculae of various shapes and dimensions and form a rosette. They are

broadest toward the upper part (middle and apex), resembling both parents. The sessile, maculate cauline leaf is narrow-acuminate, lanceolate and sheaths the stem, resembling *Dactylorhiza cordigera*. The flower bracts are longer than the flowers and purple-pigmented, resembling *Dactylorhiza majalis*. The inflorescence is an elongated, lax raceme, similar in shape and no of flowers to *Dactylorhiza cordigera* parent. The flowers are medium-sized, purple-coloured, intermediate between parental species. The flat, roundish labellum with a white base resembles in shape *Dactylorhiza majalis*, with its lateral lobes scalloped, slightly reflexed upwards. The median lobe is roundish, mildly protruding, another feature inherited from *Dactylorhiza majalis*. Labellar markings also follow *Dactylorhiza majalis*, forming circular loops extended on the lateral lobes. Lateral sepals spread upwards, similar to *Dactylorhiza majalis*. The anther is intermediate between the parents.

Nothopopulation counts: 3-4 individuals/hybrids, flowering each year.

Herbarium voucher specimen: Deposited at the Herbarium of the University of Agronomic Sciences and Veterinary Medicine of Bucharest, N.E. Anghelescu, **USAMVB–barcode 40108**.

Proposed conservation status: Endangered (EN).

× *Dactylocamptis suzana* N. Anghelescu, L. Balogh & Mih. Balogh, N. Kigyossy, 2024 **nothosp. nov.** (Figure 5).

Hybrid formula: *Anacamptis coriophora* subsp. *coriophora* × *Dactylorhiza maculata*

Discovered: 31 May 2020, Nora E. Anghelescu

Locus classicus: Prahova County, private property.

Flowering time: May-June.

Native to: Romania, Endemic (En).

Description: The stem (epigeal), 16 cm, is erect, spindly (resembling *Anacamptis coriophora*), vivid green, with purple apical pigmentation (resembling *Dactylorhiza maculata*). The two basal leaves are narrow, acuminate, vivid green, without pigmentation, a characteristic inherited from *Anacamptis coriophora*. The two cauline unspotted leaves are sessile, vivid green, acuminate, sheathing the stem. The upper one is bract-like.

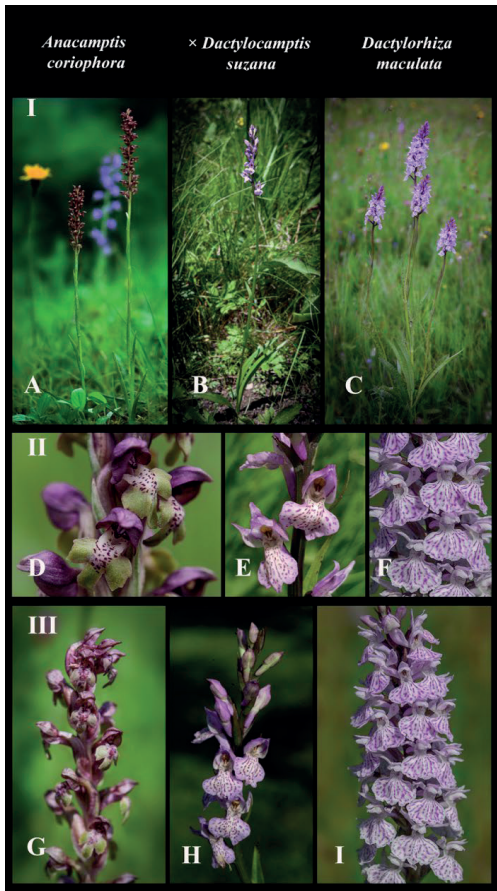


Figure 5. I. Full plants in their natural habitats; II. Individual flower detail; III. Inflorescences details; A., D., G. - *Anacamptis coriophora*; B., E., H. - *Dactylocampsis suzana*; C., F., I. - *Dactylorhiza maculata*. Photos © Nora E. Anghelescu, 31 May 2020, Prahova County

The inflorescence is an elongated, lax raceme similar to that of the *Anacamptis coriophora* parent. The flowers are medium-sized, light purple-pink, resembling *Dactylorhiza maculata*. The labellum is incurved, three-lobed, dark-spotted, and similar in shape to *Anacamptis coriophora*. The lateral petals and sepals form a tight hood over the gynostemium, resembling *Anacamptis coriophora*. The markings appear as interrupted dots that faintly create elongated loops, reflecting the typical labellar patterns of *Dactylorhiza maculata*. The spur is elongated and downcurved, resembling *Dactylorhiza maculata*, but remains thick, robust, and conical, similar to *Anacamptis coriophora*. The lateral petals and sepals also form a hood over the

gynostemium, as seen in *Anacamptis coriophora*.

Nothopopulation counts: 1 individual/hybrid, flowering each year.

Herbarium voucher specimen: Deposited at the Herbarium of the University of Agronomic Sciences and Veterinary Medicine of Bucharest, N.E. Anghelescu, **USAMVB–barcode 40106**.

Proposed conservation status: Endangered (EN).

× *Dactylanthera transsilvanica* L. Balogh & Mih. Balogh, N. Anghelescu, N. Kigyossy, 2024 nothosp. nov. (Figure 6).

Hybrid formula: *Dactylorhiza maculata* × *Platanthera bifolia*.

Discovered: 30 May 2020, Lori & Mihaela Balogh.

Locus classicus: Harghita County, private property.

Flowering time: May–June.

Native to: Romania, Endemic (En).

Description: The stem (epigeal), 46 cm is erect, slender/spindly, vivid green, resembling *Platanthera bifolia*, with purple apical pigmentation, resembling *Dactylorhiza maculata*. The basal leaf is vivid green, broad in the middle, with a more roundish apex, resembling *Platanthera bifolia*; the cauline leaves are narrow, acuminate, faintly pigmented, with elongated brownish maculae, a feature inherited from *Dactylorhiza maculata*. The three sessile, unspotted cauline leaves are vivid green and sheath the stem; the upper one is narrow-acuminate, bract-shaped. The inflorescence is an elongated, cylindrical, medium-dense raceme similar to that of the *Platanthera bifolia* parent. The flowers are medium-sized and light pink, intermediate between the two parental species, with a pale/whitish colour reminiscent of *Platanthera bifolia*. The labellum is very faintly three-lobed, with a more prominent, elongated, and downward-pointing median lobe resembling that of the *Platanthera bifolia* parent. The lateral sepals spread laterally, similar to both parental species. The median sepal is more elongated and, along with the narrower acuminate lateral petals, forms a loose hood over the gynostemium, resembling more *Platanthera bifolia* parent. The anther is more elongated than that of *Dactylorhiza maculata*, thus resembling

Platanthera bifolia. The pair of pollinia is light pinkish, resembling in shape and colour that of *Dactylorhiza maculata*. The spur is robust, cylindrical and more elongated, reminiscent of *Platanthera bifolia*, however, slightly downward pointing, resembling *Dactylorhiza maculata*. Overall, the hybrids' stems and leaves phenotypically resemble the *Dactylorhiza maculata* parent more closely, with significant resemblances to *Platanthera bifolia* in the inflorescences.

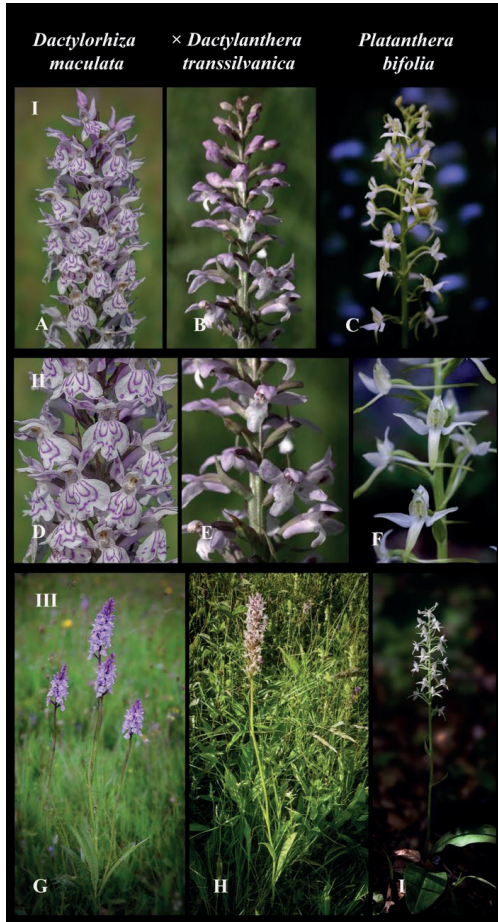


Figure 6. I. Inflorescences details; II. Individual flower detail; III. Full plants in their natural habitats; A., D., G. - *Dactylorhiza maculata*; B., E., H. - *x Dactylanthera transsilvanica*; C., F., I. - *Platanthera bifolia*. Photos © Lori & Mihaela Balogh, 30 May 2020, Harghita County

Nothopopulation counts: 1 individual/hybrid, flowering each year.

Herbarium voucher specimen: Deposited at the Herbarium of the University of Agronomic Sciences and Veterinary Medicine of Bucharest, N.E. Angheliescu, **USAMVB-barcode 40110**.

Proposed conservation status: Endangered (EN).

***Gymnadenia* × *montana* N.Angheliescu, L.Balogh, M.Balogh & N.Kigyossy, 2024 nothosp. nov.** (Figure 7).



Figure 7. I. Full plants in their natural habitats; II. Inflorescences details; III. Individual flower detail; A., D., G. - *Nigritella nigra* subsp. *bucegiana* (Hedren et al., 2022); B., E., H. - *Gymnadenia* × *montana*; C., F., I. - *Gymnadenia winkeliana* (Angheliescu et al., 2024). Photos © Nora E. Angheliescu, 29 June 2023, Prahova County

Hybrid formula: *Gymnadenia winkeliana* × *Nigritella nigra* subsp. *bucegiana*.

Discovered: 29 June 2023, Nora E. Angheliescu.

Locus classicus: Bucegi Natural Park ROSCI0013.

Flowering time: June-July.

Native to: Romania, Endemic (En).

Description: The stem (epigeal), 8-12(15) cm is erect, vivid green, with no purple apical pigmentation. 6-10 basal leaves, grass-like, vivid-green; 3-5 cauline unspotted leaves, sessile, vivid green, acuminate, sheath the stem. The roundish, hemispherical-shaped inflorescence (resembling *Gymnadenia winkeliana* parent) is a dense raceme, resembling both parents of the former subgenus

Nigritella. The flowers exhibit an overall, nearly uniform color, blending whitish-pink tones from *Gymnadenia winkeliana* and uniformly dark red hues from *Nigritella nigra* subsp. *bucegiana*, shifted toward violet (bluish) purple. The labellum presents a broader and larger shape than *Gymnadenia winkeliana*, acting as an intermediate form between the parental species, with a tendency towards the features of *Nigritella nigra* subsp. *bucegiana*. The diameter of the labellar opening or tunnel, created by the lateral labellar lobes, is somewhat wider than that of *Gymnadenia winkeliana*, closely resembling the broader and more elongated labellar opening of the *Nigritella nigra* subsp. *bucegiana* parent. The tepals are slightly narrower and more elongated, displaying an intermediate size between the two parental species. The distinctive parallel venation, marked by two parallel veins on the upper side of the lateral petals, is a trait inherited from the *Nigritella nigra* subsp. *bucegiana* parent, absent in *Gymnadenia winkeliana*.

Nothopopulation counts: 4-5 individuals/hybrids, flowering each year.

Herbarium voucher specimen: Deposited at the Herbarium of the University of Agronomic Sciences and Veterinary Medicine of Bucharest, N.E. Anghelescu, **USAMVB–barcode 40103**.

Proposed conservation status: Endangered (EN).

Epipactis × *romanensis* R. Maalouf, N. Anghelescu, L. Balogh & Mih. Balogh, N. Kigyossy, 2024 nothosubsp. nov. (Figure 8).

Hybrid formula: *Epipactis atrorubens* × *Epipactis distans*.

Discovered: 12 July 2022, Ramy Maalouf.

Locus classicus: Bucegi Natural Park ROSCI0013.

Flowering time: June–July.

Native to: Romania, Endemic (En).

Description: Stem (epigeal) measures 56 cm, erect, and green. It has one basal acuminate leaf and 7–8 unspotted, sessile cauline leaves that are green with parallel venation, ovoid-elongate shape, and acuminate tips, resembling *Epipactis atrorubens*. The phyllotaxis, which refers to the arrangement of leaves on the plant stem (Anghelescu et al., 2023), is alternate, similar to *Epipactis distans*.

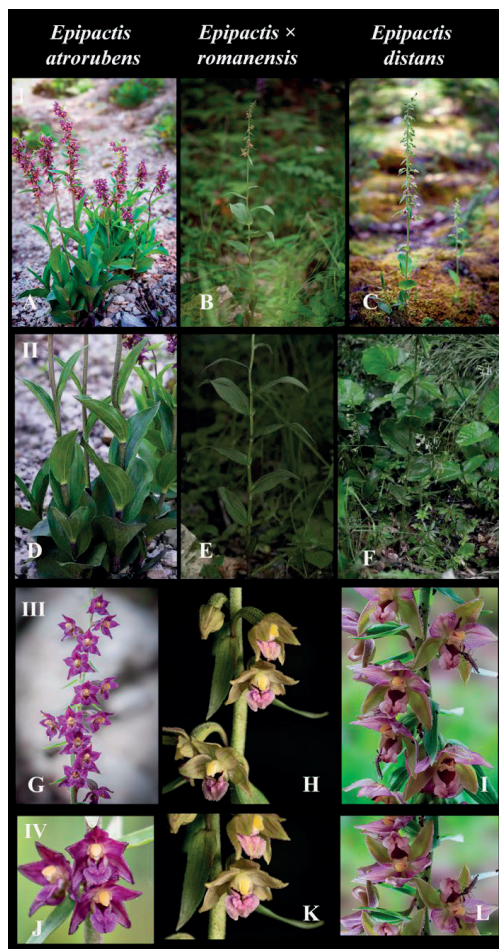


Figure 8. I. Full plants in their natural habitats; II. Details of individual phyllotaxis; III. Inflorescences details; IV. Individual flower detail. A., D., G. J. - *Epipactis atrorubens*; B., E., H. K. - *Epipactis* × *romanensis*; C., F., I. J. - *Epipactis distans*. Photos © Nora E. Anghelescu, 29 June 2022–2024, Prahova County

The inflorescence is a lax raceme, producing fewer flowers - 12–15 medium-sized blooms that resemble those of *Epipactis distans*. The flower colour is an intermediate shade between the parental species. The sepals and lateral petals are rhomboidal-acuminate, greenish with a light pink tint, and centrally green-veined, resembling *Epipactis distans*. The labellum is divided into three segments: the base, or hypochile, contains nectar similar to both parental species; the middle part, or mesochile, has deeply incurved lateral walls, is narrower than that of *Epipactis atrorubens* and is more similar to *Epipactis distans*; the apical part, or epichile, is heart-

shaped, pinkish-green, reflecting the deep red colour inherited from *Epipactis atrorubens*. The labellum features central calli with shapes intermediate between those of the parent species, specifically wrinkled and resembling more closely those of *Epipactis atrorubens*. The flower bracts are lanceolate-elongate, akin to *Epipactis distans*. The flower pedicel and ovary are tinged purple and covered in trichomes (glandular hairs), primarily a characteristic inherited from the *Epipactis atrorubens* parent.

Nothopopulation counts: 2-3 individuals/hybrids, flowering each year.

Herbarium voucher specimen: Deposited at the Herbarium of the University of Agriculture and Veterinary Medicine, Bucharest, N.E. Anghelescu, **USAMVB–barcode 40107**.

Proposed conservation status: Endangered (EN).

***Ophrys* × *minuticauda* nothosubsp. *cornuta* R. Maalouf, N. Anghelescu, L. Balogh & Mih. Balogh, N. Kigyossy, 2024 nothosubsp. *nov.* (Figure 9).**



Figure 9. I. Full plants in their natural habitats; II. Inflorescences details; III. Individual flower detail - lateral view; A., D., G. - *Ophrys apifera*; B., E., H. - *Ophrys* × *minuticauda*; C., F., I. - *Ophrys scolopax* subsp. *cornuta*. Photos © Nora E. Anghelescu, 16 June 2023, Bucegi Natural Park ROSCI0013, Prahova County

Hybrid formula: *Ophrys apifera* × *Ophrys scolopax* subsp. *cornuta*.

Discovered: 5 June 2021, Ramy Maalouf.

Locus classicus: Bucegi Natural Park ROSCI0013.

Flowering time: May-June.

Native to: Romania, Endemic (En).

Description: Stem (epigeal), 25-30 cm, erect, vivid green. 1 basal leaf, sheathing the stem, 3-5 cauline leaves, unspotted, sessile, green, acuminate (resembling *Ophrys apifera*). Inflorescence lax raceme, less floriferous, 4-5 medium-sized flowers, resembling *Ophrys apifera*, smaller than *Ophrys scolopax* ssp. *cornuta*. Sepals light-pink, centrally green veined, resembling *Ophrys apifera* (Anghelescu et al., 2021c). Lateral petals are greenish, villous (resembling *Ophrys apifera*, triangular and equal to those of *Ophrys scolopax* subsp. *cornuta*). Labellum three-lobed, median lobe slightly elongated, speculum bluish, resembling *Ophrys scolopax* subsp. *cornuta*. Lateral lobes are pointed, conical-elongated, shorter, and resemble *Ophrys scolopax* subsp. *cornuta*. Basal field orange-brown, wider, resembling *Ophrys apifera*. Appendix oblique to the labellum, intermediate between parents. Pollinia with shorter caudicle, overhanging resembling *Ophrys apifera* (correction of the original taxonomical description in Anghelescu et al., 2023, pg. 422).

Nothopopulation counts: 1 individual/hybrid, flowering each year.

Herbarium voucher specimen: Deposited at the Herbarium of the Botanical Garden Bucharest, N.E. Anghelescu, barcode: **BUC 410370**.

Proposed conservation status: Endangered (EN).

CONCLUSIONS

This study highlights the intricate complexities and ecological significance of the newly discovered nothospecies within the orchid hybrid populations. Through detailed morphological analysis and comprehensive examination of their biological characteristics, we have provided essential insights into their unique adaptations and ecological requirements. The findings not only contribute to the understanding of hybrid dynamics within the

orchid family but also emphasize the necessity for ongoing conservation efforts to protect these rare taxa. As we advocate for their formal recognition in the Romanian flora, we must prioritize the preservation of their habitats and ensure that future research continues to explore the rich biodiversity these hybrids represent. By fostering awareness and protective measures, we can safeguard the future of these remarkable and vulnerable orchid hybrids for generations to come. In conclusion, the recognition of these hybrids as new additions to the Romanian flora underscores the importance of conservation efforts aimed at protecting their habitats and promoting further research. These findings not only contribute to our understanding of hybridization processes but also maintain the ecological balance and genetic diversity within the region's ecosystems. As ongoing investigations continue to unveil the complexities of orchid hybridization, they lead the way for a deeper appreciation of Romania's rich plant heritage.

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