

FIRST RECORD OF *FESTUCA AMETHYSTINA* L. FROM THE TRANSYLVANIAN BASIN (ROMANIA)

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Abstract: Here we report the first find of a rare grass *Festuca amethystina* L. in the Transylvanian Basin. The species was found at Dealul Furcilor, a hill that is part of SCI Movilele de la Păucea near the city of Mediaș, in tall-forb vegetation classified as *Trollio europaei-Clematidetum recti* association. Using flow cytometry we detected tetraploid ploidy level of this population, corresponding to the ploidy of populations in middle altitudes of the Brașov Depression and contrasting with the diploid ploidy level of the high-montane populations in Eastern and Southern Carpathians. We show that sites of tetraploid populations share many species of dry-mesic steppe meadows and subcontinental open-canopy oak forests. Co-occurrence of *F. amethystina* and other species with disjunct distributions suggest relict character of this vegetation, probably representing remnants of ancient forest-steppe ecosystem.

Keywords: genome size, plant distribution, ploidy level, relict, tall-forb vegetation, taxonomy, Transylvanian Basin, *Trifolio-Geranietea sanguinei*, vegetation ecology, vegetation history

Introduction

Festuca amethystina L. is a grass of open and semi-open habitats with a highly uneven distribution within its range. It is broadly distributed in the montane and subalpine habitats of the Alps, Carpathians and Balkan mountain ranges, while it is scattered or rare elsewhere, particularly in the lowlands and hilly landscapes of Central Europe [16, 22]. It is classified as a rare species in Romania [27] and included in Red Lists also in other countries, e.g. Hungary [17], Czech Republic [10] and Poland [14]. It prefers mesic to dry-mesic grasslands and open canopy forests (mostly *Pinus*- or *Quercus*-dominated) on base-rich to moderately base-rich soils, often developed over limestones, other calcareous sediments or ultramafic rocks.

In Romania, *F. amethystina* occurs mainly in the high-montane and subalpine grasslands of the Eastern and Southern Carpathians [1, 5, 8]. Recently it was reported also from submontane open-canopy oak forests of the Brașov Depression [12]. While the high-montane and subalpine populations in Romania seem to be diploid, those in lower altitudes are tetraploid [28]. Although such ploidy differentiation in *Festuca* usually reflects different taxonomic status [e.g. 15], taxonomic consequences of ploidy variation in *F. amethystina*, particularly the relation to the four recognized subspecies [18, 21], as yet remain unresolved [28].

Here we report the first find of *F. amethystina* in the Transylvanian Basin, together with data on its ploidy and accompanying vegetation. We discuss several related taxonomic and ecological issues in the broad geographical context.

Material and Methods

Study area

Dealul Furcilor (435 m a.s.l.) is situated between the villages Păucea, Blăjel and Romanești, 7 km north of the city of Mediaș (Sibiu county), in the south-central part of the Transylvanian Basin. The hill is a part of a treeless ridge above the valley of Păucea stream, a tributary of Târnava Mare river. The ridge is formed of calcareous claystones of Neogene age. On northern slopes, the relief is diversified by landslips. The hill is protected within the Site of Community Importance (SCI) Movilele de la Păucea [23, 38].

Morphology and ploidy level

Festuca amethystina can be easily identified due to the absent awns and the specific anatomical structure of the tiller leaves, particularly the organization of sub-epidermal sclerenchyma strands, forming 3–5 ribs along the whole length of tiller leaf sides. The latter character can be observed even in the field on well-preserved desiccated or dead leaves. Nevertheless, we checked the anatomical structure also in transverse cross sections performed on the middle parts of several leaves, using an Olympus SZ-X7 binocular microscope ($\times 50$ magnification). We further checked the morphological characters given as characteristic for the subspecies *F. amethystina* subsp. *amethystina* and subsp. *orientalis* in the latest Romanian key [33].

For the estimation of ploidy level, relative genome size was measured approximatively from the dried leaves of the herbarium voucher using flow cytometry with DAPI fluorescence dye. The procedure and concentration of chemicals follow Šmarda [37], with the exception of using *Bellis perennis* L. (Asteraceae) as the internal genome size standard. Measurements were done with a Partec ML flow cytometer equipped with UV LED diode (Sysmex).

Vegetation composition

Vegetation data were recorded in a 4×4 m plot using standard phytosociological methodology, with the extended Braun-Blanquet scale used for species cover-abundance estimation [7]. Topsoil was sampled in several places within the relevé, and pH and conductivity of a mixed sample were measured in aquatic solution using a Hach HQ40d multi meter.

Taxonomic concepts and nomenclature of vascular plant taxa follow Euro+Med PlantBase [9], except for *Avenula praeusta* and *Campanula moravica*, for which the concept of Sârbu et al. (2013) was used, and the following species complexes (aggregates): *Dianthus carthusianorum* agg. (*D. carthusianorum*, *D. giganteiformis*, *D. giganteus*), *Ranunculus polyanthemos* agg. (*R. nemorosus*, *R. polyanthemos*), *Veronica chamaedrys* agg. (*V. chamaedrys*, *V. vindobonensis*) and *Vicia cracca* agg. (*V. cracca*, *V. tenuifolia*).

Results

Festuca amethystina was found in an abandoned tall-forb-rich steppe meadow on the northern slope of Dealul Furcilor near the city of Mediaș during phytosociological sampling on 24 June 2018 (Fig. 1). The relevé and data on site conditions are provided below. Herbarium vouchers from the plant are deposited in the Herbarium of the Masaryk University (BRNU) in Brno. The plant is tufted, without extravaginal shoots, and its tiller leaves are scabrous throughout their length. The *F. amethystina*/*Bellis perennis* nuclei fluorescence ratio was 2.154 (mean of two measurements) which corresponds to tetraploids based on our unpublished

measurements of diploid and tetraploid *F. amethystina* populations from the Central Europe, Ukraine and the Balkans.

Romania, Mediaş region, SCI Movilele de la Păucea, Dealul Furcilor, latitude 46°13'35.1"N, longitude 24°20'24.2"E (WGS-84), altitude 400 m a.s.l., plot size 16 m², slope inclination 35°, slope aspect 60°, soil pH 6.21, soil conductivity 254 µS/cm, cover of herb layer 70%, cover of moss layer 1%. Authors: J. Roleček, P. Dřevojan, M. Hájek & P. Hájková.



Fig. 1: *Festuca amethystina* at its newly found site at Dealul Furcilor in the Transylvanian Basin. Tall plants of *Clematis recta* dominate the stand, and *Geranium sanguineum*, *Mercurialis ovata* and *Sanguisorba officinalis* are also visible.

Herb layer: *Clematis recta* 3, *Carex montana* 2b, *C. filiformis* 2m, *Festuca amethystina* 2m, *Avenula praeusta* 2m, *Stachys officinalis* 2m, *Brachypodium pinnatum* 1, *Ferulago sylvatica* 1, *Mercurialis ovata* 1, *Adenophora liliifolia* +, *Agrimonia eupatoria* +, *Agrostis capillaris* +, *A. vinealis* +, *Allium oleraceum* +, *Anthericum ramosum* +, *Anthoxanthum odoratum* +, *Aristolochia lutea* +, *Avenula pubescens* +, *Campanula glomerata* +, *C. moravica* +, *C. persicifolia* +, *C. rapunculoides* +, *Cruciata glabra* +, *Cytisus albus* +, *Dactylis glomerata* +, *Dianthus carthusianorum* agg. +, *Elytrigia intermedia* +, *Euphorbia angulata* +, *Festuca rubra*

+, *Filipendula vulgaris* +, *Galium verum* +, *Geranium sanguineum* +, *Gladiolus imbricatus* +, *Hypochoeris maculata* +, *Lathyrus pannonicus* subsp. *collinus* +, *L. transsilvanicus* +, *Lilium martagon* +, *Luzula campestris* +, *Peucedanum oreoselinum* +, *Plantago media* +, *Potentilla alba* +, *Primula veris* +, *Prunella grandiflora* +, *Ranunculus polyanthemos* agg. +, *Rumex acetosa* +, *Sanguisorba officinalis* +, *Schedonorus pratensis* +, *Securigera varia* +, *Stellaria graminea* +, *Tanacetum corymbosum* +, *Teucrium chamaedrys* +, *Thesium linophyllum* +, *Trifolium alpestre* +, *T. montanum* +, *T. pannonicum* +, *Trollius europaeus* +, *Veronica chamaedrys* agg. +, *Vicia cracca* agg. +, *Vincetoxicum hirsutiflorum* +, *Carlina acaulis* r, *Centaurea oxylepis* r, *Fragaria viridis* r, *Helleborus purpurascens* r, *Laserpitium latifolium* r, *Thalictrum aquilegifolium* r; *Crataegus* sp. r.

Discussion

Ploidy level

Tetraploid ploidy level of the *F. amethystina* population at Dealul Furcilor is in agreement with the results of Rewicz et al. (2018), suggesting altitudinal differentiation between high-montane diploids and lower-altitude tetraploids in Romania. The cited study indicates that different ploidy levels cannot be simply associated with any of the four recognized subspecies based on the identification characters currently used. However, the correspondence between the distribution patterns of diploids and *F. amethystina* subsp. *orientalis* (Eastern and Southern Carpathians) [21] suggests that Romanian diploids may be probably identified with this subspecies. This view is also supported by diploid ploidy level recorded in a population on Blyznytysya Mt. in Ukrainian Eastern Carpathians (Bednarska & Šmarda, unpubl.), close to the type locality of *F. amethystina* subsp. *orientalis* (Svydovets) [18].

Though the morphology of the plant corresponds clearly to *F. amethystina* subsp. *amethystina* according to Sârbu et al. (2013), the correct subspecific name for the reported tetraploid population is unclear. This is due to (i) the probable diploid ploidy level of this (nominata) subspecies: the lectotype was designated from Western Alps [3], where diploids of *F. amethystina* are reported by Litardière (1950) and Rewicz et al. (2018); and (ii) unclear taxonomic status of a tetraploid subspecies *F. amethystina* subsp. *ritschlii*, reported from Central Europe [21], but presently accepted only in Poland [24] and Germany [13].

Vegetation

Our record is in line with a growing body of evidence that the current distribution of *F. amethystina* reflects not only its habitat requirements, but also long-term history of its sites. It has been hypothesized long ago that low-altitude sites of *F. amethystina* have refugial origin and the species is a glacial or early Holocene relict [35, 36]. Indeed, perhaps all new records of *F. amethystina* in low and middle altitudes of Central Europe [e.g. 12, 19, 26, 32] come from similar habitats: either subcontinental open-canopy oak forests (*Potentillo albae-Quercetum* association and closely related communities) or semi-dry and intermittently wet grasslands (*Cirsio-Brachypodium* and *Molinion* alliances). That is habitats whose species-pool is believed to be inherited from glacial or early Holocene hemiboreal forest-steppe communities [4, 11, 29]. This hypothesis is supported by the occurrence of species with disjunct distributional ranges in the preserved stands: at Dealul Furcilor, *Adenophora liliifolia* and *Lathyrus transsilvanicus* can be mentioned besides *F. amethystina*.

Although tall-forb-rich steppe is a habitat as yet unreported for *F. amethystina*, it has many features in common with other low-altitude sites of the species. For example, Indreica (2007) reports ten species shared with our relevé from subcontinental open-canopy oak forests in the Braşov Depression: *Brachypodium pinnatum* (dominant), *Campanula glomerata*, *C. persicifolia*, *Carex montana* (dominant), *Euphorbia angulata*, *Potentilla alba*, *Primula veris*, *Stachys officinalis*, *Veronica chamaedrys* agg. and *Vincetoxicum hirundinaria*. Almost all these species are characteristic of forest-steppe habitats such as dry-mesic grasslands, forest fringes and open-canopy forests, usually on deep, base-rich soils. As noted before [2, 30, 34], tall-forb-rich steppe meadows are a regular component of forest-steppe mosaics at relatively wet and productive sites (northern slopes, terrain depressions), where conditions are suitable also for species more abundant at higher altitudes or latitudes (at Dealul Furcilor, besides *F. amethystina* also *Aconitum variegatum* or *Trollius europaeus*). In the past such vegetation was possibly more common in the montane or northern forest-steppe.

Remarkably, the phytosociological association *Trollio europaei-Clematidetum recti*, encompassing tall-forb-rich steppe meadows, was firstly described exactly from Dealul Furcilor [38]. The authors put the association into the *Calthion* alliance and *Molinio-Arrhenatheretea* class, which is clearly erroneous. Based on its physiognomy (dominance of tall forest-steppe herbs) and the occurrence of many diagnostic species (e.g. *Anthericum ramosum*, *Brachypodium pinnatum*, *Carex montana*, *Campanula persicifolia*, *Clematis recta*, *Euphorbia angulata*, *Ferulago sylvatica*, *Geranium sanguineum*, *Laserpitium latifolium*, *Lathyrus pannonicus* subsp. *collinus*, *L. transsilvanicus*, *Peucedanum oreoselinum*, *Tanacetum corymbosum*, *Trifolium alpestre*) [25], we classify it within the *Trifolio-Geranietea sanguinei* class. According to our experience, similar vegetation is scattered over a large area in Transylvania and beyond [30, 31]. Several syntaxa names introduced later (particularly *Clematido recti-Laserpitietum latifolii* Schneider 1984 and *Clematido-Avenuletum praeustae pubescentis* subass. *clematido recti-laserpitiosum latifolii* Bădărău 2005) may be considered synonymous with *Trollio-Clematidetum*. Unlike Coldea et al. (2012), we consider the latter name validly published, oldest and therefore correct name for the respective vegetation type.

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**PRIMA SEMNALARE PENTRU *FESTUCA AMETHYSTINA* L.
ÎN BAZINUL TRANSILVANIEI (ROMÂNIA)**

(Rezumat)

Menționăm pentru prima dată apariția unei specii rare de graminee, *Festuca amethystina* L. în bazinul Transilvaniei. Specia a fost găsită pe Dealul Furcilor, care face parte integrată din SCI Movilele de la Paucea, lângă Mediaș, în vegetație de ierburi înalte ce aparține asociației *Trollio europaei-Clematidetum recti*. Cu ajutorul citometriei de flux s-a detectat un nivel de tetraploidie în cadrul populației, similar populațiilor de altitudine medie din Depresiunea Brașovului și deosebit de diploidia populațiilor de altitudine înaltă din estul și sudul Carpaților. Demonstrăm că siturile cu populații tetraploide adăpostesc multe specii de pajisti stepice mediu-uscate și de păduri de stejar subcontinentale. Apariția speciei *Festuca amethystina* în acest context și a altor specii cu areal disjunct sugerează caracterul relictar al acestui tip de vegetație, reprezentând probabil reminiscențe ale ecosistemului străvechi de păduri stepice.