In vitro establishment, propagation and conservation of the rare and endangered moss halophyte *Hennediella heimii*

Milorad Vujičić[1], Beáta Papp[2], Aneta Sabovljević[1], Erzsébet Szurdoki[2] and Marko Sabovljević[1]

[1]Institute of Botany and Botanical Garden, Faculty of Biology, Univesristy of Belgrade, Takovska 43, 11000 Belgrade, Serbia
[2] Department of Botany, Hungarian Natural History Museum, H–1476 Budapest, Pf. 222, Hungary

BBGB

Abstract

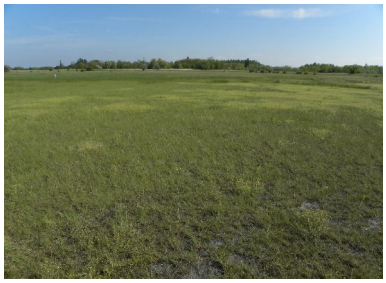
Hennediella heimii

The rare moss *Hennediella heimii* (Hedw.) R.H. Zander (Pottiaceae) is a small plant of bipolar distribution. It inhabits salt soils, which are temporarily under water. It grows among other vascular halophytic plants and is one of the seldom moss species growing in salty environment. The plant material was collected at: Hungary, Bács-Kiskun County, Szappanszék at Fülöpháza village, saline-alkali area, 46°53'11,1"N, 19°25'35,3"E, 115 m a.s.l., 11.05.2010.
With aim to established stable axenical culture and large-scale micro-propagation and plants productions for the purposes of introduction, reintroduction and further research on biology of this species, we have optimized growth conditions by studying environmental condition variations and the effect of exogenous plant growth regulators.
The axenic culture was established after surface sterilization of sporophytes and the spreading of spores over the nutritive media. Spores germinate in small percentage (up to 17%). After germination the protonema remained small, but dense. The fully developed gametophytes has been achieved in BCD medium, a year after germinations at low light short day regimes.

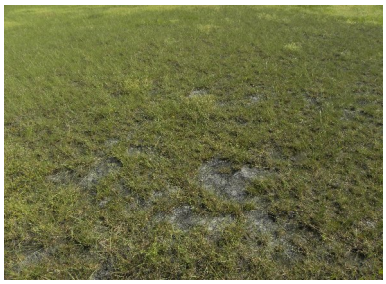
Hennediella is a somewhat puzzling genus which had been previously included in various genera, such as *Desmatodon, Pottia, Syntrichia*, and *Tortula*. It was re-defined by Zander (1993) who distinguished 20 species, with a distributional center mainly in the southern hemisphere. Today 15 species with additional two varieties within the genus *Hennediella* are recognized. *H. heimii* is widely but rather rare moss species, scattered in its distribution and restricted to short life span in harsh environment. The geographical distribution includes various continets but the species is rare to find due to specific ecology and its biological features. It is distinguished as being a xeric moss species that grows in habitats with changing water availability and is, therefore, exposed to alternating cycles of desiccation and rehydratation. Another interesting features of this species includes acclimatisation to high light intensity by building up highly effective non-photochemical quenching systems, salt tolerance, and supporting high temperature amplitude.

It is threatened in many European countries: Vulnerable in Swiss and Czeck Republic, Critically endangered in Spain, Belgium, Austria and Germany, Endangered in Hungary, Extinct in Italy. It is known from the literature data also from Bosnia, Romania, Serbia and Slovenia.

This species grows in the meadowes or sparse halophytic grasslands in the lowlands on soils with highsalt contentc, periodically inundated (in spring) and dominated by *Puccinellia*. Bryophyte species composition and, particularly, percentage cover depended mainly on the texture of the soils, the duration of wetting and only at the most extremely salty sites on the degree of salinity. *Entosthodon hungaricus* and *Pottia heimii* were the only species that were restricted to saline soils.



Saline-alkali area at Bács-Kiskun County, Hungary



With an aim to produce genetically diverse axenic cultures of *H.heimii* the way of establishing whole plants from the variety of spores has been studied. The problems in establishing moss culture has been elaborated elsewhere (e.g. Rowntree et al. 2011). The sterilization process were tested with few sterilization substances in various concentration (NaOCl, ethanol and NaDCC 5, 7, 10, 13%; each). The spore germination were tested from dry versus young and fresh mature capsules. The germination were evaluated on various media (MS; MS/2; BCD w/wo sugar), light (long day/short day) and temperature (18°C vs. 25±2°C) conditions. Once the spore germinated the influences of media and condition applied on primary protonema were estimated in view of achieving the highest percentage of bud formations and gametophore development (index of multiplication). The behavior of secondary protonema developed on separated gametophore were estimated for the purpose of massive micropropagation.

In vitro establishment, propagation and conservation

A – after succesfull sterilization, spore gerimante and interesstinglly, the protonema form callous like structure.

B – threatment with exogenous added plant growth regulators (PGR), induces buds on primary protonemal filaments

C – PGR also act on shot formation on secondary developed protonemal filaments

D, E, F – fully developed plants of *H. heimii*