

## **Hydrochemical Characterization of A Stand of the Threatened Endemic *Isoëtes malinverniana***

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## SHORTER NOTES

**Hydrochemical Characterization of A Stand of the Threatened Endemic *Isoëtes malinverniana*.**—*Isoëtes malinverniana* Ces. & De Not. is an aquatic quillwort endemic to Northern Italy that, as with many other quillworts, is facing drastic changes in its habitat (Wen *et al.*, J. Freshwater Ecol. 18:361–367. 2003.). *Isoëtes malinverniana* grows in running water canals used for rice field water supply (usually with *Ranunculion fluitantis* vegetation), but in the past it probably occurred in natural streams generated by springs and minor river branches, characterized by oligotrophic waters (Abeli *et al.*, Aquatic Conserv: Mar. Freshw. Ecosyst. 22:66–73. 2012). During the last forty years the distribution range of *I. malinverniana* has been rapidly decreasing as a consequence of changes in the rice cultivation practices in Northern Italy. Particularly, the use of herbicides and fertilizers, the regimentation of water courses with water removal in winter, and the mechanical re-profiling of the canals are the major threats to the species (Barni *et al.*, Aquat. Bot. 107:39–46. 2013). Although the species is protected at European and national levels, its conservation status is critical (Bilz *et al.*, European Red List of Vascular Plants. Publications Office of the European Union, Luxembourg. 2011; Rossi *et al.*, Lista Rossa della Flora italiana. 1. Policy Species e altre specie minacciate. Comitato Italiano IUCN e MATTM. 2013), and urgent conservation actions are needed to stop the decline of the species, by reinforcing some extant populations and/or reintroducing new populations within the historical range. The major problem with respect to reintroduction actions is the fact that the original habitat of *I. malinverniana* has been greatly modified. This reduces the probability of successful translocations and also affects the possibility to study the real ecological requirements of the species. However, a site of *I. malinverniana* with about 30 plants discovered a few years ago in a natural stream in the Ticino river basin at La Sforzesca near Vigevano (voucher specimen in PAV) still has many of the characteristics of the original habitat. Here we have analyzed the ion concentrations of surface water and, for the first time, sandy sediment pore water along a 30 m long transect crossing the population. Pore water samples were collected in three points along the transect at about 10 m from each other, while a single surface water sample was collected in the middle of the transect. Sediment pore water samples were collected at a depth of about 10 cm using ceramic cups (Eijkelkamp Agrisearch Equipment, Giesbeek, the Netherlands), connected to 100% vacuum PVC syringes (50 ml) by means of a PVC tube, according to Van Der Welle *et al.* (Freshwater Biol. 52:434–447. 2007). For each water sample, pH, electrical conductivity and temperature were immediately recorded, while ion concentrations were analyzed at the laboratory of the Radboud University (Nijmegen, The Netherlands).

Surface water pH, electrical conductivity and temperature were identical among the three samples (6.8; 271  $\mu\text{S}/\text{cm}$ ; 11°C, respectively). This pH was

lower than in populations within channels used for rice field water supply (Barni *et al.*, 2013; Table 1), but higher than the mean pH values found in Dutch and Norwegian populations of *I. lacustris* and *I. echinospora* (Table 1). The pH of the sediment pore water was remarkably high, reaching a mean value of  $8.7 \pm 0.9$ , which also results in very low pore water carbon dioxide concentrations. Mean pH values in Dutch and Norwegian *Isoëtes* stands are around 6, whereas pore water carbon dioxide concentrations are typically (much) higher than surface water concentrations (Table 1). Being able to take up carbon dioxide by the roots, this situation provides a competitive advantage for isoëtid species compared to non-isoëtid species, which lack this ability (Smolders *et al.*, *Aquat. Bot.* 73:325–350. 2002). Therefore *Isoëtes* species thrive very well in soft water lakes and streams where carbon availability in the water layer is low and species dependent on carbon uptake from the water layer are unable to grow. The high calcium, magnesium and bicarbonate concentrations of the water layer and sediment pore water indicate that the water in the stand from the Ticino river basin is relatively well-buffered compared to the Dutch and the Norwegian *Isoëtes* stands, which were found in weakly buffered soft waters. The Ticino population is probably dependent on the uptake of carbon dioxide from the water layer where carbon dioxide concentrations are much higher than in the sediment pore water. Pedersen *et al.* (*J. Exp. Bot.* 62:4691–4700. 2011.) showed the potential gas exchange via the leaves to be substantial for *I. australis*, although the resistance to gas exchange was up to three times higher than for roots. The uptake of CO<sub>2</sub> via the roots may have further lowered CO<sub>2</sub> concentrations and indirectly increased the pH of the sediment pore water in the Ticino stand.

Regarding the surface water, the concentration of phosphate, total-P and ammonia are at the lower end of the ranges found for Dutch and SW Norwegian *Isoëtes* stands and are also lower than the mean values found for other *I. malinverniana* stands (Table 1). However, the nitrate concentration is unnaturally high (Table 1), suggesting water nutrient enrichment probably due to the presence of rice and cornfields just a few hundred meters upstream from the population.

High concentration of nitrate was also evident in sediment pore water. Although for the parameters analyzed in the sediment pore water, it was not possible to make a comparison with other stands of *I. malinverniana*, pore water nitrate was very high compared with values measured in Dutch and Norwegian *Isoëtes* stands (Table 1) and values reported for isoëtid lakes in Spain (Catalan *et al.*, *Hydrobiologia* 274:17–27. 1994; Gacia *et al.*, *J. Limnol.* 68:25–36. 2009) and in Scandinavia (Vestergaard & Sand-Jensen, *Aquat. Bot.* 67:85–107. 2000). *Isoëtes* species generally grow on mineral, usually sandy, sediments with low oxygen consumption rates and actively maintain the sediment in an oxidized state by leaking oxygen via the roots (Pedersen *et al.*, 2011). Due to the oxidized conditions nitrate, iron and sulphate reduction, are normally not important in isoëtid stands. This results in low iron concentrations in sediment pore water, as iron is not mobilized by iron reduction (Table 1), which also results in a low mobility of phosphorus, which is

TABLE 1. Ion concentration for surface water and sediment pore water of a stand of *Isoëtes malinverniana*. Values of surface water concentration from Barni *et al.*, 2013 and unpublished data for three *Isoëtes* stands from the Netherlands (A.J.P. Smolders) and different *Isoëtes* stands from 9 lakes in SW Norway, sampled in 1995 and 2010 (E.C.H.E.T. Lucassen) are shown for comparison. Data are expressed in  $\mu\text{mol/L}$ .

Surface water	<i>Barni et al. (2013)</i>	<i>This study</i>	(Neth, 2005) <i>Isoëtes</i>	(Norw, 1995) <i>Isoëtes</i>	(Norw, 2010) <i>Isoëtes</i>
	<i>Isoëtes malinverniana</i>				
pH	7,6	See text	5,8 (0,2)	5,7 (0,7)	6,2 (0,6)
HCO <sub>3</sub>		1303	35,3 (7,5)	12,0 (25,5)	113 (168)
CO <sub>2</sub>		454	123,3 (13,7)	76,4 (16,3)	84,1 (88,8)
P		0,4	3,0 (1,9)	0,8 (0,7)	0,4 (0,3)
PO <sub>4</sub>	3,55	0,13	0,4 (0,2)	0,3 (0,3)	0,3 (0,3)
NO <sub>3</sub>	26,6	149	16,1 (13,7)	11,1 (5,1)	9,2 (7,2)
NH <sub>4</sub>	5,5	1,6	12,7 (8,3)	3,8 (3,4)	2,7 (3,7)
Ca	520	819	246 (145)	38,3 (20,3)	35,9 (16,1)
Mg		277	81,5 (36,5)	23,8 (6,2)	23,5 (5,0)
Na		316	413 (191)	145 (41,0)	182 (25,0)
Cl		216	613 (290)	166 (41,6)	172 (28,5)
K		54,4	110 (57,8)	10,4 (11,0)	13,8 (13,0)
Fe		0,1	6,5 (5,9)	1,2 (0,9)	1,8 (2,2)
Mn		0,02	2,2 (2,3)	0,2 (0,2)	0,1 (0,1)
SO <sub>4</sub>		302	107 (35,5)	31,9 (8,0)	22,6 (1,9)
Si		181	36,6 (30,8)	15,7 (5,8)	6,2 (7,4)
Al		0,02	13,1 (8,6)	7,3 (3,2)	1,8 (0,6)
n		1	3	8	9

  

Pore water	<i>Barni et al. (2013)</i>	<i>This study</i>	(Neth, 2005) <i>Isoëtes</i>	(Norw, 1995) <i>Isoëtes</i>	(Norw, 2010) <i>Isoëtes</i>
	<i>Isoëtes malinverniana</i>				
pH		8,7 (0,9)	6,0 (0,1)	5,9 (1,0)	6,1 (0,5)
HCO <sub>3</sub>		3069 (569)	395 (189)	175 (224)	192 (175)
CO <sub>2</sub>		16 (3)	730 (115)	336 (306)	275 (177)
P		9,6 (1,1)	3,1 (2,3)	0,6 (0,4)	1,5 (0,8)
NO <sub>3</sub>		119 (21,9)	10,3 (12,6)	11,8 (12,7)	15,5 (17,1)
NH <sub>4</sub>		3,5 (3,4)	29,4 (13,6)	3,9 (2,2)	1,7 (2,2)
Ca		1021 (95)	337 (169)	102 (61,4)	146 (127)
Mg		816 (326)	126 (54,7)	73,8 (55,5)	60,8 (34,8)
Na		1988 (802)	435 (192)	214 (78)	228 (122)
Cl		1780 (643)	435 (192)	192 (94)	197 (121)
K		330 (186)	96,7 (31,4)	13,7 (19,3)	31,0 (25,0)
Fe		0,2 (0,2)	8,1 (4,9)	2,6 (2,2)	0,9 (1,3)
Mn		1,3 (1,6)	3,1 (0,8)	1,2 (1,8)	0,4 (0,7)
SO <sub>4</sub>		344 (16,4)	200 (233)	70,9 (51,6)	69,4 (17,4)
Si		246 (25,5)	83,5 (55,6)	98,3 (68,7)	125,9 (90,7)
Al		0,2 (0,1)	21,9 (18,2)	10,3 (7,7)	2,9 (6,6)
n		3	3	30	20

efficiently bound to oxidized iron(hydr)oxides (Smolders *et al.*, 2002). Also the observation that at our location nitrate and sulphate concentrations do not differ between water layer and pore water suggest the lack of microbiological consumption of nitrate or sulphate in the sediment, indicating oxidative conditions in the sediment.

The chemical characterization of the *I. malinverniana* habitat shows that the species is growing on an oxidized sediment with a relatively low availability of phosphorus. The water layer and sediment are relatively well buffered and characterized by a high pH and a very low availability of carbon dioxide in the sediment pore water. Although the surface water phosphorus concentrations are still low, the nitrate enrichment due to the intensive agricultural activity of the area is evident even in a site apparently less impacted than other stands. As a consequence even a temporary increase of the phosphorus availability in the water layer might easily lead to an excessive growth of algae. This poses serious threats for the conservation of this endemic species for which a suitable site for translocation is at the moment unavailable. Flowing water may strongly benefit the species under more eutrophic conditions because in flowing water algae are less likely to become dominant and uptake of CO<sub>2</sub> from the water layer is facilitated. Nevertheless increased nutrient levels in the water layer will at least lead to the growth of epiphytic algae as has been observed in many of the remaining *I. malinverniana* populations (e.g. Arborio, Vigevano), which may depress growth by shading and by depletion of inorganic carbon and nutrients at the leaf surface.—T. ABELI (e-mail: thomas.abeli.it@gmail.com), S. ORSENIGO and N. M. G. ARDENGHI, DSTA, Department of Earth and Environmental Sciences, University of Pavia, via S. Epifanio 14, 27100, Pavia, Italy, E.C.H.E.T. LUCASSEN and A.J.P. SMOLDERS, Department of Aquatic Ecology and Environmental Biology, Radboud University, Heyendaalseweg 135, 6525 AJ Nijmegen, The Netherlands.