

## Long-term monitoring of an invasion process: the case of an isolated small wetland on a Mediterranean Island, second stage: toward a complete restoration

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**Abstract:** In the present article, the results of the first-stage of monitoring, following restoration works on a small Mediterranean wetland (Lake Stagnone, Capraia Island, Tuscan Archipelago), are reported. The recent spread of *Typha* and *Phragmites* in the lake changed diversity and composition of the plant communities. Nine years after their first monitoring (2009), a rarefaction of hydrophytes and small helophytes of conservation interest was detected. In 2010, the restoration started with the aim to remove (or at least reduce) the populations of the large, expansive helophytes. In 2012, the first post-actions monitoring were carried out using the same methods as previously, analysing the plant presence/absence and their cover value recorded in the same 15 plots selected in 2000 and 2009. The rise and fall of the populations of the various flora and vegetation types during this invasion process and the following restoration were statistically analysed. One year following the restoration, some recovery (replacement) had occurred of autochthonous hydrophytes and small helophytes. Many of these species are of conservation interest. Some aquatic plants, present on the site until the more or less recent past, were once more recorded. Given the rapid recovery of populations of many autochthonous species, the results are reasonably encouraging, rendering planned reintroductions unnecessary at the moment. On the other hand, because of the short time elapsed since restoration, the current community structure cannot in any way be considered an “equilibrium” one. Continued and regular monitoring is required to allow the reestablishment of the large expansive helophytes populations.

**Key words:** conservation; hydrophytes; NMDS; plant diversity; SDR

### Introduction

Many authors have noticed that aquatic ecosystems, such as wetlands, lakes, ponds, etc., are key in hosting high biodiversity at both regional and global scales (e.g., Denny 1994; Bedford et al. 2001; De Meester et al. 2005; Dudgeon et al. 2006). Nowadays, however, these habitats are particularly under threat from a wide range of anthropogenic factors (Davis & Froend 1999; Bedford et al. 2001; Dudgeon et al. 2006).

The loss or the large modification of a freshwater habitat can be particularly damaging in “water-stress” regions such as in the Mediterranean area, where the seasonal and yearly variation can be very large (Acreman 2000). In this ecological context, human activity can have major direct and indirect impacts on the hydrological dynamics necessary for the well being or survival of wetland areas (see Médail et al. 1998; Acreman 2000).

Among the threat factors, an important role is often played by the invasion of alien or native plants, leading not only to a reduction of plant and animal diversity

but also to a modification of the ecological features of the invaded habitats (Zedler & Kercher 2004).

Some large helophytes such as *Typha* sp. pl. and *Phragmites australis* appear particularly able to colonize wetland ecosystems under these conditions, with consequential loss of native biodiversity and a major alteration of ecosystem function including nutrient cycling and hydrology (see Lenssen et al. 2000; Green & Galatowitsch 2001; Ehrenfeld 2003; Zedler & Kercher 2004; Bowles & Jones 2006; Boers et al. 2006; Lorens 2006; Tulbure et al. 2007).

Invasion by the large helophytes *Typha angustifolia*, *T. latifolia* and *Phragmites australis* represented the source of the rapid changes first reported by Foggi et al. (2011) in the small lake “Lo Stagnone” on Capraia Island in the north Tyrrhenian Sea (Tuscany, Italy). Even if the term “invasive” was, sometimes, applied also to native species (Valéry et al. 2004; Tighe et al. 2009), it is mostly formally associated to alien species (Richardson et al. 2000; Pyšek et al. 2004). In this paper, we indicate with the term “expansive” the large helophytes, native in Italy but unknown at the Stagnone Lake un-

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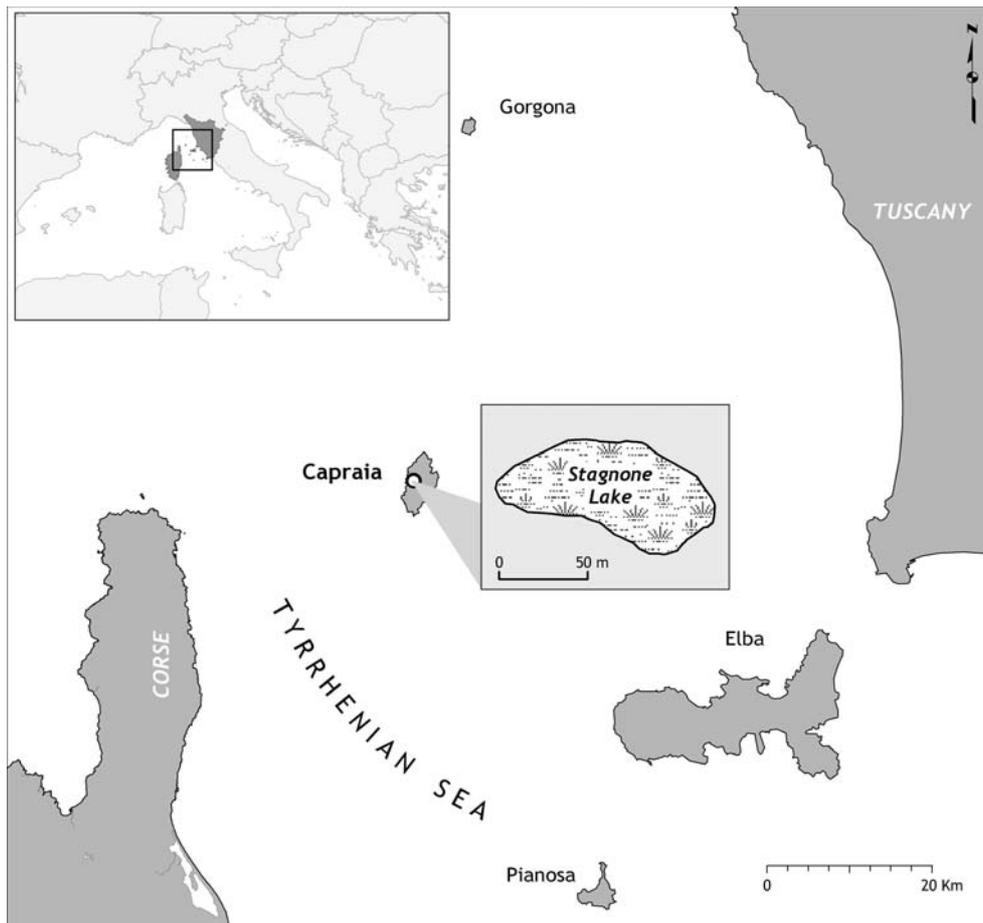


Fig. 1. The study area.

til few years ago (Foggi et al. 2011); furthermore, the term “autochthonous” is used to indicate the species (hydrophytes and small helophytes) which were present in the lake before the arrival and the stabilization of the expansive helophytes.

Lake Stagnone is the only one permanent wetland in the Tuscan Archipelago: on the other hand, other wet ecosystems in this area are represented mostly by small, temporary pools and streams or disturbed and relict wet areas.

The invasion of Lake Stagnone by cattail and common reed rapidly led to the formation of dense stands dominated by these helophytes and to the detriment of the open-water areas occupied by autochthonous hydrophytes such as *Ranunculus peltatus* subsp. *baudotii*, *Myriophyllum alterniflorum*, and small helophytes like *Eleocharis palustris* and *Baldellia ranunculoides*. The open areas of the lake have been drastically reduced in less than twenty years (Foggi et al. 2011). Even if it is not certain how the large helophytes have been arrived to the Stagnone lake, one of the possible causes could have been the opening of the Capraia Island to tourists, especially birdwatchers after the closure of the prison (Foggi et al. 1999; Lastrucci et al. 2010). This could have favored the accidental arrival of seeds of these plants to the lake, maybe involuntarily transported from other continental wetlands. Despite the identifying of the processes leading to the degrada-

tion of the studied ecosystems constitutes an important steps in restoration (Hobbs & Norton 1996; Hobbs & Harris 2001), the concrete risk of local extinction of the above reported species has urgently required the re-establishment of open-water spaces to allow the recovery of the autochthonous species and vegetation of the lake. The restoration project has necessarily involved the removal of these expansive helophytes. The project was financed by the Tuscan Regional Administration and was carried out under the direction of the National Park of the Tuscan Archipelago. The first phase of the work was started in June 2010 and was completed in September 2011.

Aim of this paper is to evaluate the short-term responses of Lake Stagnone’s plant communities following the first phase of the restoration, by comparing the plants recorded in the plots in the early and later (i.e. more advanced) phases of the helophyte invasion. In order to evidence the changing trend, new statistical analyses of the invasion have been made, additionally functional for extending the analyses previously presented in a first report (Foggi et al. 2011).

## Material and methods

### Study area

Capraia Island belongs to the Tuscan Archipelago (north central Mediterranean), 53 km from the Italian coast and

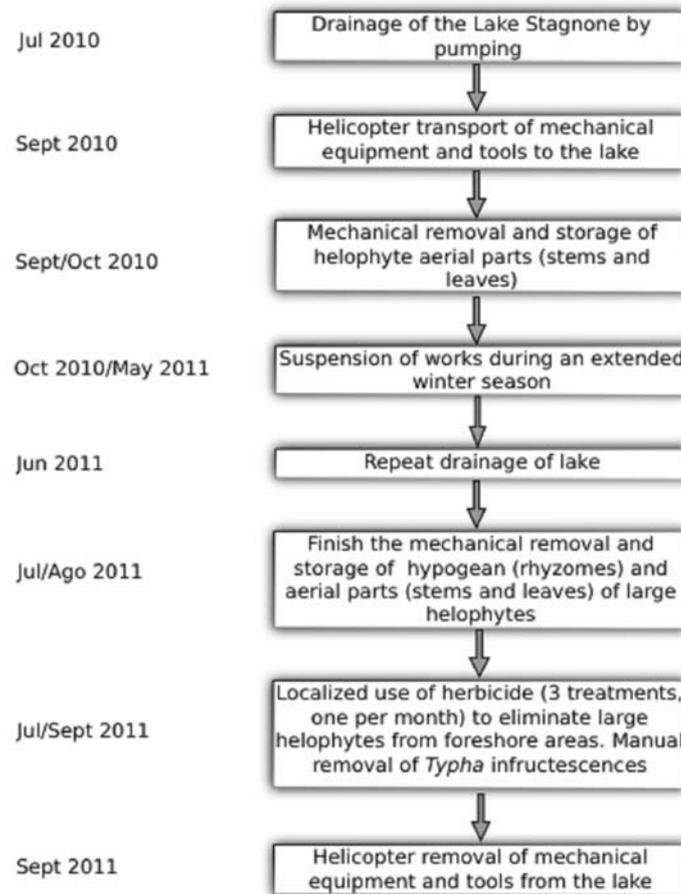


Fig. 2. Flowchart of restoration works.

26 km from the island of Corsica (Fig. 1). The island has an area of 19.72 km<sup>2</sup>; the maximum length is about 8 km and the maximum width is about 4 km (Foggi & Grigioni 1999). Capraia is entirely constituted of volcanic rocks; several alluvial areas are locally present. Climate is typically Mediterranean with hot, dry summers and wet winters (Foggi et al. 2011). Stagnone Lake is situated on Capraia at an altitude of 330 m a.s.l., within a small saddle. The deepest part of the lake is about 140 cm and its water originates exclusively from local precipitation. For further, more detailed environmental information see Foggi et al. (2011).

#### Technical details of the works

Technical operations faced many practical difficulties linked to the following factors: (1) Problematic site access by heavy machinery such as excavators and trucks; (2) Almost continuous presence of water; (3) The expansive helophytes were dispersed over the entire water surface, with the total number of stems (ramets) being estimated at around one million (roughly 200 to 300 stems m<sup>-2</sup>); (4) The small area available for the temporary, local storage of the material removed; (5) The high sensitivity/fragility of the ecosystem. The various phases of operations are summarised in Fig. 2. All these factors forced us to employ small machinery, easily transportable to/from the site by helicopter.

The removal of helophytes was carried out mostly with mechanical methods. After the drainage of the lake, all the aerial and the under-ground parts (rhizomes) found in the first 10-20 cm of soils were removed by mean of a mini-excavator. The whole resultant plant material was put into burlap sacks and stored.

The use of chemical herbicides, such as those based on glyphosate, was carefully assessed. Although the technical and scientific literature contains a number of cases of successful control/eradication of cattails and common reed by employing these products (Messersmith et al. 1992; Mozdzer et al. 2008), they might also have undesirable effects on non-target species (Marrs et al. 1989; Gardner & Grue 1996) and a number of groups of animal, such as amphibian species results particularly vulnerable (Mann & Bidwell 1999; Relyea 2005). In addition to plant species of conservation interest, a large population of the Tyrrhenian tree frog (*Hyla sarda*) is also here present. This Sardinian-Corsican endemic species is listed in Annex IV of the European Habitat Directive 92/43, in Annex A2 of the Tuscan Regional law 56/2000 on biodiversity, and is also included in the Red List of Italian fauna (Bulgarini et al. 1998). For all these reasons, chemical weed control was limited to a few marginal and non-submerged areas of the foreshore using a foliar spray formulation based on the herbicides glyphosate (3%), picloram (3%) and ammonium sulphate (2%). The chemical treatment was monthly repeated from June to September 2011. Along the shoreline, manual removal of cattail seed heads (about 150) was carried out to decrease the risk of spread in the following season. Manual removal of the new resprouts of *Typha* and *Phragmites* is provided each year until the summer of 2015.

#### Sampling design for floristic study

The field sampling followed the method reported by Foggi et al. (2011).

Table 1. List of recorded species. For each species the growth form, the occurrence percentage, the ISA pval and the class of maximum indicator value are reported. Those species having special conservation interest are printed in bold. HA, annual herb; HB, biennial herb; HP, perennial herb; WF, woody frutex; lHe, large helophyte; sHe, small helophyte; Hyr, rhizophyte; Hyp, pleustophyte; maxcls, class (year) in which each species assumes its maximum indicator value; indval, indicator value for each species; pval, probability of obtaining as high an indicator values as observed over 1000 iterations.

Species	Abbreviation	Growth form	Occurrence %			ISA		
			2000	2009	2012	maxcls	indval	pval
<i>Agrostis stolonifera</i> L.	Agr_sto	HP	13.3	20.0	33.3	2009	0.146	0.576
<i>Alisma plantago aquatica</i> L.	Ali_pla	sHe	46.7	40.0	20.0	2000	0.435	0.032
<i>Anagallis arvensis</i> L.	Ana_arv	HA	0.0	6.7	6.7	2012	0.060	1
<b><i>Baldellia ranunculoides</i></b> (L.) Parl.	Bal_ran	sHe	6.7	6.7	53.3	2012	0.527	0.002
<i>Briza minor</i> L.	Bri_min	HA	0.0	0.0	6.7	2012	0.067	1
<i>Callitriche stagnalis</i> Scop.	Cal_sta	Hyr	6.7	0.0	0.0	2000	0.067	1
<i>Carex divisa</i> Huds.	Car_div	HP	13.3	26.7	13.3	2009	0.117	0.626
<i>Chara braunii</i> C.C. Gmelin	Cha_bra	Hyr	0.0	0.0	20.0	2012	0.200	0.092
<i>Cynodon dactylon</i> (L.) Pers.	Cyn_dac	HP	0.0	6.7	6.7	2009	0.061	1
<i>Cyperus badius</i> Desf.	Cyp_bad	HP	0.0	26.7	20.0	2012	0.165	0.292
<i>Daucus carota</i> L.	Dau_car	HB	0.0	0.0	6.7	2012	0.067	1
<i>Ditrichia viscosa</i> (L.) Greuter	Dit_vis	HP	6.7	20.0	6.7	2009	0.075	0.678
<b><i>Eleocharis palustris</i></b> (L.) Roem.	Schult.	Ele_pal sHe	40.0	6.7	13.3	2000	0.187	0.348
<i>Geranium columbinum</i> L.	Ger_col	HA	0.0	0.0	6.7	2012	0.067	1
<i>Geranium purpureum</i> Vill.	Ger_pur	HA	0.0	0.0	6.7	2012	0.067	1
<i>Juncus acutus</i> L.	Jun_acu	HP	0.0	6.7	6.7	2009	0.033	1
<i>Juncus articulatus</i> L.	Jun_art	HP	6.7	0.0	20.0	2012	0.109	0.311
<i>Juncus effusus</i> L.	Jun_eff	HP	46.7	46.7	33.3	2009	0.192	0.754
<i>Lemna minor</i> L.	Lem_min	Hyp	0.0	73.3	0.0	2009	0.733	0.001
<i>Lotus angustissimus</i> L.	Lot_ang	HA	0.0	0.0	6.7	2012	0.067	1
<i>Mentha pulegium</i> L.	Men_pul	HP	0.0	6.7	0.0	2009	0.067	1
<b><i>Myriophyllum alterniflorum</i></b> DC.	Myr_alt	Hyr	33.3	13.3	60.0	2012	0.454	0.019
<i>Oenanthe pimpinelloides</i> L.	Oen_pim	HP	6.7	6.7	6.7	2000	0.022	1
<i>Phragmites australis</i> (Cav.) Trin. ex Steud.	Phr_aus	lHe	0.0	6.7	6.7	2009	0.047	1
<i>Polypogon subspathaceus</i> Req.	Pol_sub	HA	0.0	0.0	6.7	2012	0.067	1
<i>Potamogeton crispus</i> L.	Pot_cri	Hyr	6.7	0.0	40.0	2012	0.359	0.012
<b><i>Ranunculus peltatus</i></b> Schrank subsp. <i>baudotii</i> (Godr.) C.D.K. Cook	Ran_bau	Hyr	60.0	20.0	66.7	2000	0.398	0.028
<i>Ranunculus sardous</i> Crantz	Ran_sar	HA	6.7	6.7	0.0	2000	0.061	1
<i>Rubus ulmifolius</i> Schott	Rub_ulm	WF	13.3	20.0	40.0	2012	0.178	0.417
<i>Rumex acetosella</i> L.	Rum_ace	HP	0.0	6.7	6.7	2009	0.061	1
<i>Rumex conglomeratus</i> Murray	Rum_con	HP	26.7	26.7	13.3	2009	0.076	0.99
<i>Sonchus asper</i> (L.) Hill	Son_asp	HA	0.0	6.7	6.7	2009	0.061	1
<i>Trifolium nigrescens</i> Viv.	Tri_nig	HA	0.0	6.7	0.0	2009	0.067	1
<i>Trifolium repens</i> L.	Tri_rep	HP	0.0	0.0	6.7	2012	0.067	1
<i>Typha angustifolia</i> L.	Typ_ang	lHe	26.7	53.3	13.3	2009	0.398	0.069
<i>Typha latifolia</i> L.	Typ_lat	lHe	33.3	40.0	0.0	2009	0.309	0.073
<i>Veronica anagallis-aquatica</i> L.	Ver_ana	sHe	6.7	0.0	0.0	2000	0.067	1

The study area was rasterised with a 5×5 m grid and 15 squares were selected at random during the year 2000. In the centre of each selected square, a 1×1 m plot was marked out and georeferenced, and the entire flora was recorded. Cover values were estimated in the field using the Braun-Blanquet scale (1932). The original Braun-Blanquet scale values were transformed as follows: 5 = 87.5%, 4 = 62.5%, 3 = 37.5%, 2 = 15%, 1 = 2.5%, + = 1%, and r = 0.1% (see also Wild et al. 2004; Bimová et al. 2004). The same plots were visited during the month of May in 2000, 2009 and 2012. Species nomenclature follows Conti et al. (2005; 2007). For each species, the growth form observed in the survey area was reported following Den Hartog & Segal (1964) and Arrigoni (1996). Small and large size helophytes have been distinguished and counted separately for monitoring.

#### Data analyses

Changes in the pattern of species composition were evaluated using non-metric, multidimensional scaling (NMDS) and the analysis was carried out with *R* software (R Development Core Team 2013) using the *vegan* package (Oksanen

et al. 2013). The Bray-Curtis similarity index was used as a distance measure. The same package was used for the Multi-response permutation procedure (MRPP) with the aim of testing compositional differences between the three floristic assembly recorded over the three years of sampling. The Bray-Curtis similarity index was used as a distance measure. Separation between groups was calculated as the chance-corrected, within-group agreement (A) and the p-value was used to evaluate the how likely an observed difference was due to chance.

Indicator Species Analysis (ISA) was adopted to determine how strongly each species was associated with the year of sampling (Dufrêne & Legendre 1997); ISA was performed with *R* software using the *labdsv* package (Roberts 2012). An SDR-simplex analysis (Podani & Schmera 2011) was carried out to explore species-richness and species-diversity patterns. The analysis results are displayed in a ternary plot where each square symbol represents a pair of sites. The presence/absence data matrixes (one for each year) were analysed using the MS Windows application SDR-Simplex developed by Podani. The ternary plots were drawn using



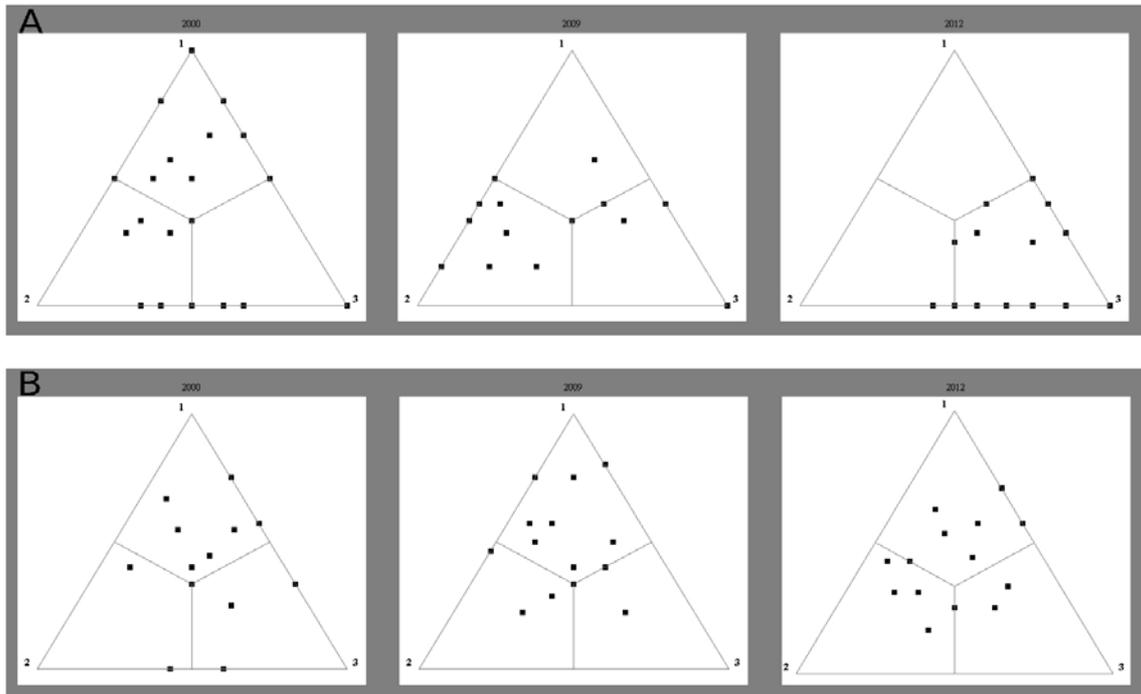


Fig. 5. SDR simplex triplots for: (A) The aquatic, and (B) The shore communities in the three observation periods. The vertices of the triplots represent: 1) Species replacement, 2) Richness difference, and 3) Similarity.

in the first phase (from 2000 to 2009) but returned to a higher value after the eradication. On the other hand, the large helophytes reached their highest value in 2009. The NMDS analysis (Fig. 4; stress = 0.068) showed a clear separation between the plots on the shorelines (located mainly in the upper-left quadrant) and those of the mainly-submerged areas (located in the other three quadrants). The aquatic plots of 2000 fall almost exclusively in the low-right quadrant, while those of 2012 are distributed between the upper and lower-right quadrants. Plots dominated by the genus *Typha* (almost all in 2009) are concentrated in the lower-left quadrant (Fig. 4).

The indicator-species analysis (ISA) shown in Table 1 emphasised that six species are statistically significant ( $p < 0.05$ ) for the three time-steps considered. For the 2000 plots – *Alisma plantago-aquatica* and *Ranunculus peltatus* subsp. *baudotii*; for the 2009 plots – *Lemna minor*; for the 2012 plots *Baldellia ranunculoides*, *Myriophyllum alterniflorum* and *Potamogeton crispus*. In Table 1, the year of monitoring in which each species assumes its maximum value of indication is also reported. The results of the SDR analyses of the aquatic plots were reported in Table 2 and, graphically, in Fig 5A. The plots of 2000 gave the highest contribution to Species Replacement (39.5%), those of 2009 and 2012 gave the highest contribution to Similarity (respectively 36.2% and 59.2%). Similarity and Species Replacement of the 2012 plots were significantly different from 2000 and 2009 ones. For all foreshore plots (Table 2; Fig. 5B) the highest value was for Species Replacement. The differences between the R, D and S values of the plots in the three different years were not statistically significant.

Table 2. Percentage contributions from the SDR-simplex analysis. Significantly different values ( $p < 0.05$ ; Kruskal-Wallis test) among S, R and D for each habitat type are marked with the letters (a, b).

	Aquatic plots		
	2000	2009	2012
Similarity (S)	31.8a	36.2a	59.2b
Species replacement (R)	39.5a	29.4a	14.5b
Richness difference (D)	28.7a	34.4a	26.3a
	Shore plots		
	2000	2009	2012
Similarity (S)	36.7a	23.4a	28a
Species replacement (R)	37.9a	48.5a	41.5a
Richness difference (D)	25.4a	28.1a	30.4a

## Discussion

In the recent years, the rapid spread of *Typha angustifolia*, *T. latifolia* and, more recently, *Phragmites australis*, have together caused a rapid change in the vegetation of the Stagnone, leading to an almost complete closure of open-water areas, habitat of the autochthonous hydrophytes (Foggi et al. 2011). The surveys carried out in 2012, after the eradication of the large helophytes, showed several interesting novelties.

Even if some species, such as *Veronica anagallis-aquatica* and *Callitriche stagnalis*, observed here until 2000 and not recorded since, the creation of open-water areas suitable for hydrophytes and the drastic reduc-

tion in the population of the large helophytes, has allowed *Potamogeton crispus* to reappear. This species has not been recorded since the cattails and the reeds became established but is now fairly widespread, with its occurrence rising from 6.7% in 2000 to 40% in 2012. Furthermore, two rhizophytes historically most abundant in the lake (see Moris & De Notaris 1839; Foggi & Grigioni 1999; Foggi et al. 2011), *Ranunculus peltatus* subsp. *baudotii* and *Myriophyllum alterniflorum*, have benefited from the restoration activity, with a higher occurrence in 2012 than in 2000.

*Baldellia ranunculoides* has also been favoured from the operations. Its presence in 2009 was reduced to a few individuals located at the edge of the lake, whereas in 2012 it was widely distributed in the lake. These three species are perhaps the most interesting from a conservation point of view, being all included in the Red List of Tuscany (Conti et al. 1997). Moreover, the other plant present in regional attention lists for nature conservation, *Eleocharis palustris*, has increased its occurrence in comparison with 2009. Of the large helophytes responsible for the invasion, only *Typha latifolia* was absent from the 2012 plots. *Typha angustifolia* was still present but in reduced numbers. Treatments appear to have had fewer effects on *Phragmites australis*, since its population remained substantially unchanged from 2009 until 2012. Nevertheless, its community, earlier dense and almost monospecific, afterward consisted of a mosaic of sparse plants intermingled with autochthonous hydrophytes.

*Lemna minor* was absent from the 2012 plots: as stated by some authors (Whyte et al. 2008; Foggi et al. 2011), its presence was related to the occurrence of the large size helophytes; the decreasingly sheltered environment created by the removal of emergent vegetation could have had an important role in the disappearance of *Lemna minor* from the 2012 plots. It must be noted, however, that a few individuals of *Lemna* and several plants of *Typha latifolia* were observed in marginal areas of the lake in 2012.

Eight new species were observed in 2012. Among these, it is worth noting the re-discovery of *Chara braunii*. This species is fairly rare in Italy nowadays (Bazzichelli & Abdelahad 2009) and its presence in Lake Stagnone was recorded for the last time over a hundred years ago (Béguinot & Formiggini 1908). Several authors have stressed the high capacity of dormant oospores of some of these algae to remain quiescent for many years and then to germinate when conditions become favourable (Bonis & Grillas 2002). Moreover, the presence of algae of the genus *Chara* is often associated with the early stages of re-colonisation after an intervention (Lorens 2006).

Floristic changes in the plant community of 2012, compared to that of 2009, are also indicated in the NMDS and ISA results. Overall, two different ecological situations can be distinguished.

I) The first concerns the true aquatic coenoses in the central area of the lake, where the operation were concentrated. Here, at greater or lesser depths, the more

direct interest was in the spread of cattails and subsequently reeds. The analyses show that, from floristic and functional points of view, the intervention restored the plant communities to a condition more similar to that recorded in 2000, rather than to that in 2009. The plots recorded in 2012 are dominated mainly by rhizophytes, rather than by the large helophytes of 2009. The situation with rhizophytes now widespread throughout the lake is probably similar to the pre-invasion situation, or at least to the physiognomy of plant communities recorded up to 1995 (Foggi et al. 2011). The significant recovery of aquatic communities after restoration is shown also by SDR analysis. The transition from “mosaic” communities with the invaders *Typha* and *Phragmites* (and *Lemna minor*) mixed with the autochthonous hydrophytes and small helophytes towards a “primitive” strictly aquatic community after the removal of the expansive species, resulted in a decrease of Species Replacement and in a increase of Similarity.

II) The second ecological situation concerns the plots on the foreshore, which were less intensely affected by the restoration work. Here, the NMDS shows less difference between the 2009 and 2012 plots. This aspect is confirmed also by SDR analysis, that showed no statistically significant differences among the different communities.

However, in 2012, several annual species appeared at the edges of the lake i.e.: *Geranium purpureum*, *G. columbinum*, *Polypogon subspathaceum*, *Briza minor* and *Lotus angustissimus*. Many of these, show a pioneering behavior and their establishment in the area could be due to the disturbance during the restoration actions.

Plants and seeds of autochthonous species of highest conservation value from the site were collected and stored in the Seeds-Bank of the Pisa's Botanical Garden in anticipation of the possible need for their reintroduction. Studies on the *ex-situ* germination of seeds of *Ranunculus peltatus* subsp. *baudotii* from Stagnone were carried out with good results, enabling such reintroduction (Carta et al. 2012). The first stage results of the monitoring are encouraging, given the rapid recovery of natural populations of *R. peltatus* subsp. *baudotii* and the reoccurrence of other hydrophytes and small helophytes disappeared during the spread of the large helophytes; thus, their reintroduction may not be necessary.

The current community structure should not be considered an a new “equilibrium” of the all Stagnone-ecosystem, given the relatively short period passed by the actions. It is more likely that the plant communities will further evolve into more mature arrangements in which the relations of presence and cover of the various species will adjust naturally.

It is noted that not all individuals of *Typha* and especially of *Phragmites* have been removed and some further actions are planned for the coming years. Therefore, Lake Stagnone's vegetation should continue to be monitored regularly, to allow quick identification of new trends of species expansion and enabling prompt inter-

vention to prevent reestablishment of the large helophyte populations. Furthermore, as above mentioned, it will be important to in deep investigated the causes of the arrival of the expansive species in the lake to get an effective and lasting restoration. According to the current hypothesis, one of the actions that maybe important, could consist of an information campaign aimed at the training of a responsible tourism.

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