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Short communication

The rare aquatic angiosperm *Elatine gussonei* (Elatinaceae) is more widely distributed than previously thought

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ABSTRACT

Elatine gussonei (Sommier) Brullo et al. is a rare freshwater plant previously assumed to be endemic to Lampedusa and the Maltase archipelago. Taxonomic uncertainties within the *Elatine* genus may have caused *E. gussonei* populations to be overlooked in the Mediterranean region. To clarify the distribution of *E. gussonei*, we reviewed *Elatine* specimens from 12 herbaria and conducted eight field surveys in Mediterranean countries. Through our herbarium review we documented previously unknown occurrences of *E. gussonei* from Portugal, Spain, France, Algeria, Egypt, Cyprus and Israel. Additionally, in field studies we found populations from Cyprus, Morocco and Spain. *Elatine gussonei* is therefore more widespread than previously assumed, although the species still has a scattered distribution around the Mediterranean and many of the records are old. We found intact *E. gussonei* seeds in the faeces of migratory Greylag Geese (*Anser anser*) collected in southern Spain, suggesting that *E. gussonei* has a high capacity for long distance dispersal.

1. Introduction

Plant species of the class Isoëto-Nanojuncetea are rare elements of the European flora, and many are threatened by extinction (Bilz et al., 2011; Lukács et al., 2013). The genus *Elatine* L. suffers from taxonomic uncertainty due to a high degree of phenotypic plasticity (Mason, 1956; Molnár et al., 2015), and the distribution and habitat requirements of different species are unclear. The different species are distributed mostly in the northern hemisphere, predominantly in areas with a temperate climate, but the size of their known geographical range varies from extensive (e.g. *E. alsinastrum* L.) to very restricted (e.g. *E. gussonei* (Sommier) Brullo et al.). The clarification of taxonomic status and the reliable assessment of distribution is required for the effective conservation of any species (Primack, 2010). Mapping distributions also allows us to answer fundamental questions in ecology, such as patterns of abundance, rarity, or species richness at different spatial scales (Baselga et al., 2012).

Elatine gussonei (Fig. S2) was first described as *E. hydropiper* var. *gussonei* Sommier based on the shape of the seed and the length of flower pedicels (Sommier, 1908), and was later classified as a separate species (Brullo et al., 1988). It is rare and threatened, and was considered to be endemic to the archipelago of the central part of the Mediterranean Basin (Lampedusa and Malta; Sommier, 1908; Mifsud, 2006; Kalinka et al., 2014) until new locations were recently discovered in Sicily (Molnár et al., 2014; Minissale and Sciandrello, 2014). According to the IUCN Red List (Lansdown, 2011), the population trend of *E. gussonei* is decreasing and the total area of occupancy is unlikely to exceed 100 km^2.

As part of ongoing research into the taxonomy, biology and distribution of *Elatine* species in Europe we visited several suitable habitats and reviewed existing herbarium sheets. Here, we report new records of *E. gussonei* around the Mediterranean region which indicate a major extension of the previously recognized distribution. According to our recently published molecular phylogeny (Sramkó et al., 2016), *E.*

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Table 1
The number of reviewed sheets and the number of sheets where *Elatine gussonei* was found, sorted by the original taxon name on the labels.

Taxa name on the label	Number of reviewed sheets	Number of sheets with seeds	Number of *E. gussonei* sheets	Proportion of misidentified *E. gussonei* sheets (%)
E. macropoda	190	128	36	28
E. campylosperma (incl. *hydropiper* var. *pedunculata; hydropiper* f. *campylosperma*)	74	63	33	52
E. gussonei (incl. *hydropiper* var. *gussonei*)	6	5	5	0
E. major	8	5	2	40
E. aquatica	1	1	1	100
E. hydropiper	7	2	1	50
E. fabri	6	3	1	33
E. hardyana	1	1	0	0

gussonei is clearly a separate species. Samples from Sicily, Morocco and Spain form a monophyletic group, whereas samples from Lampedusa and Malta are clustered together with *E. macropoda* Guss. due to recent hybridisation between the two species (Sramkó et al., 2016). Unfortunately, Sramkó et al. (2016) failed to point out this hybrid lineage *E. gussonei* × *macropoda* in their paper. If this hybrid lineage is excluded, *E. gussonei* is delimited as a monophyletic entity.

The main aims of our study were 1) to clarify the distribution range of *E. gussonei* in the Mediterranean Basin and 2) to report evidence for long-distance dispersal capacity which helps to explain its scattered distribution over an extensive range.

2. Materials and methods

Distribution data of *E. gussonei* were obtained from field sampling and the examination of a total of 293 specimens (Table 1) deposited in 12 herbaria (Table S1). We studied herbarium sheets of *Elatine* taxa with opposite leaves and tetramerous flowers that were collected in the Mediterranean.

Field sampling campaigns were conducted in Morocco, Portugal, Spain, Malta, Sicily, Sardinia, Lampedusa and Cyprus. During field sampling, we visited those sites of *E. gussonei* identified from the reviewed herbarium sheets, and we searched suitable habitats (temporary and permanent ponds) for the species in surrounding areas.

Because vegetative characteristics show high overlap between *Elatine* species (Mifsud, 2006; Molnár et al., 2015), we paid particular attention to the seeds, to ensure correct identification. The main differentiation characters between the tetramerous *Elatine* species in Europe are observable in the seed curvature and the reticulation on the testa (Molnár et al., 2015) (Fig. S1). These characters were evaluated according to the identification key presented by Popiela et al. (2017). Namely, the specimens which have seeds with (80–)180–247(–347)° curvature on average, and have 17–23(–32) hexagonal pits in the middle row of the seed reticulation, were recorded as *E. gussonei*.

In late winter (12 February 2016), we collected 10 faecal samples of Greylag Goose (*Anser anser*) from Caño de Rosalimán in Doñana National Park (N 37.07513°, W 06.39077°) in southern Spain, where *E. gussonei* was recorded according to our reassessment of herbarium sheets. The mean mass of each sample was 1.99 g (range 0.69–6.53 g). Samples were closely inspected to avoid contamination from soil or plant parts before placing them in individual plastic bags and storage at 5 °C until processing. Faeces were washed in a 100 µm sieve with deionised water. Sieved contents were then inspected with a Zeiss microscope to locate seeds. After the separation, we immediately conducted germination tests by placing all seeds on moistened filter paper in Petri-dishes. The paper was regularly irrigated during the tests. The Petri-dishes were placed in a germination chamber with a 12 h:12 h photoperiod. During the light period the temperature was 22 °C and during the dark period it was 18 °C. Seeds were checked daily for germination for 30 days.

3. Results and discussion

Taxonomic and biogeographical research of phenotypically plastic species is particularly important for species of conservation importance such as *E. gussonei*. We reviewed 293 sheets of the tetramerous species of the *Elatinella* section in 12 herbaria (Table S1). For 85 sheets (29%), we could not make a reliable identification mainly due to the lack of seeds. We found *E. gussonei* in 79 sheets with seeds, but most of them (74 specimens, 94%) had been misidentified. Altogether we identified 59 localities (Table S2). 27% of reviewed herbarium sheets (38% of the sheets with seeds) were classified as *E. gussonei*. Table 1 provides the taxon-names under which *E. gussonei* had been misclassified. We detected mixed specimens (*E. gussonei* + *E. macropoda*) in two cases (Table S2).

Our study indicates that *E. gussonei* has a considerably wider geographic distribution than formerly assumed and is quite widespread around the Mediterranean (Fig. 1). Beyond the previously known distribution (Lampedusa, Malta, Sicily) we found current populations in Morocco (Ben Slimane, N 33.61388°, W 07.10129°), Spain (Casar de Cáceres, N 39.55333°, W 06.42000°) and Cyprus (Peyia, N 34.88473°, E 32.35952°; Neo Chorio, N 35.01505°, E 32.30002°), and through the revision of herbarium material, we identified previously unknown sites from Portugal, Spain, France, Algeria, Egypt, Cyprus and Israel. Since most of the herbarium data originated before 1950, the current status of *E. gussonei* in these sites should be assessed. The extent to which *E. gussonei* has been under-recorded is illustrated by the fact that it has until now been overlooked in the Doñana wetlands in SW Spain, despite the fact that this area has been extensively studied (Green et al., 2016b).

In line with previous statements (Sommier, 1908; Mifsud, 2006; Molnár et al., 2014; Minissale and Sciandrello, 2014), *E. gussonei* mainly prefers limestone rock pools (Fig. S3 A–C). Besides that, we observed the presence of *E. gussonei* along muddy lakeshores (Fig. S3 D) and in temporary pools (Fig. S3 E). Based on the labels of the herbarium sheets, *E. gussonei* also prefers marshes (Algeria, Egypt), oxbows, ditches and temporarily inundated depressions (France) (Table S2).

Extensive overlap in the distribution, ecology and morphology of *E. macropoda* (Mifsud, 2006; Popiela and Łysko, 2010) and *E. gussonei* has obviously contributed to the high frequency of misidentifications. At the same time, the single suitable distinctive feature, namely the seed morphological characters identified by Sommier (1908), have been widely ignored until now. Unfortunately, identification keys for Mediterranean *Elatine* taxa erroneously suggest that *E. macropoda* may have