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Australian acacias across eastern Adriatic – abundant but not aggressive

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Non standard abbreviations:
FH = flower head
ADD = *A. dealbata dealbata*
ADS = *A. dealbata subalpina*
Phen + no. = pheno-phase

Key words: invasive plants, phenology, distribution, Croatia

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Abstract

Background and Purpose: *Biological invasions are considered among the main factors of global changes and the identification of future invaders may help in taking effective steps to prevent their dispersal and/or establishment. For some Australian* Acacia *species, especially* Acacia dealbata *Link, an invasive status has been already described in some European countries. The aim of this work was surveying the actual spatial pattern of Australian acacias across the eastern Adriatic, and, by applying GIS techniques, to learn more about their ecology and distribution.*

Materials and Methods: *We carried on two field surveys during the flowering season of the year 2014, and recorded the locations of* Acacia *species, their flowering state, floral characteristics, and the insects present on the inflorescences. We georeferenced any plant detected, and collected soils samples under the plant crowns and in close natural or semi-natural areas.*

Results and Conclusions: *Very few species were recorded and mostly in private gardens,* A. dealbata *being the preferred ornamental. Records referring to plants grown in abandoned or unattended gardens demonstrate the potential of this species to expand if not properly controlled. In addition, we detected no plants outside gardens in natural habitats. Results on soil characteristics, with higher pH values than those usually found under trees of this genus elsewhere, may partly explain our findings. The surveys took place during a spring following an unusually warm winter: this may have influenced both phenology and pollinator activity, finally reducing observations of the latter. Descriptive statistics and MaxEnt analysis have shown the importance of the warmer areas in determining the presence of* Acacia *species. At present, acacias are not to be considered aggressive invaders along the eastern Adriatic according to our observations; they can be classified as alien, with a local trend toward becoming casual.*

INTRODUCTION

Alien invasive species are a main topic on which European Policy is recently focussing on *(1)*. Thanks to the contribution of many researchers to the EU funded project DAISIE (www.europe-aliens.org), we currently know that 6658 alien species (the highest number; at June 2014) can be found in the taxon group of the terrestrial plants, representing one of the greatest threats to the ecological and economic balance of different natural vegetation. Alien plants interact with native ones in many ways, e.g. influencing their population dynamics *(2, 3)*, their spatial distribution *(4, 5)*, and the interactions with pollinators *(6, 7)* and seed dispersers *(8)*. Global consequences of alien invasions can be hard to counteract, especially when considering that gaps and contrasting economic interests exist at the international level *(9)*. Yet the best way to address the issue of alien invasive species is expanding our knowledge on their distribution, the ecological conditions in which they grow and the interactions they establish with native species.

Acacia is a large cosmopolitan genus, able to colonise different type of habitats. Worldwide there are more than 1350 species, and it is the largest genus of woody plants in Australia with almost 1000 species *(10)*. Australian acacias show different status of invasiveness in Europe: several species display an invasive status in some Mediterranean countries, but not in others *(11, 12)*. *A. dealbata* Link., possibly the best known species and the most widespread, is native to south-eastern Australia and Tasmania and has been originally introduced to Europe as garden species since at least 1824; it was also used in forestry or land rehabilitation *(13, 14)*. Acacias prefer moist soils, but well-drained (DAISIE, http://www.europe-aliens.org/aboutDAISIE.do); however some species, as *A. dealbata*, may tolerate drier soils, strong wind, moderate frosts and snow (down to –7 °C). It is still planted for forestry, as an ornamental plant and for soil stabilization, especially in Mediterranean areas. Its growth form may be a bush or a spreading tree up to 30 m high, and it is especially appreciated for its abundant and yellow inflorescences with showy displays occurring in late winter/early spring. Lorenzo *et al*. *(15)* summarised the case of *Acacia* invasion, listing the *Acacia* species with invasive potential in Europe. *A. dealbata* is included in that list, spreading in many Mediterranean countries: Portugal, Spain, France and Italy. In an Italian invaded area with sclerophyllous native vegetation, Lazzaro *et al*. *(16)* found *A. dealbata* to show a crucial ability in changing soil conditions, strongly influencing also biotic elements, as the composition of the bacterial, fungal and understory plant communities, consistently to the results from Spain *(17)* and Portugal *(18, 19)*. First efforts on management of *A. dealbata,* by combining cutting and herbicide use, have been reported from NW Spain by Souza-Alonso *et al. (20)*. So far, no information has been reported on the invasion status of *A. dealbata* on the other side of the Mediterranean Sea, the eastern Adriatic, even though some local ecological conditions may be similar to those areas where the species has managed to express its invasion ability. *A. dealbata* has long been studied for its capacity in N_2-fixation *(19, 21)*, its invading potential in many parts of the world *(22-24)*, and it is one of the few alien plants for which some records exist on the interactions with pollinators in the invaded areas *(6)*.

The climatic conditions of the eastern Adriatic may be suitable for its spread, considering acacias grow in tropical, subtropical, and warm temperate parts of the world. This area has also been confirmed as very important for the overall native biodiversity *(25-26)*; therefore understanding the distribution pattern and the bio-ecology of (potentially) invasive species is a crucial conservation concern. The main goal of this study was to investigate acacias distribution along the Croatian coast, with special attention to *A. dealbata.* Flora Croatica Database (http://hirc.botanic.hr/fcd) reports the presence of *Acacia dealbata* Link, *A. saligna* (Labill.) H. L. Wendl. f. and *A. retinoides* Schltdl. However, these species are not indicated as invasive in Croatia. Australian *Acacia* species, alien in the Mediterranean area with varying impact on the natural vegetation, may successfully be adopted for developing trends in the arrival, spread and impacts of invasive alien species across Europe and help to unravel so far neglected barriers.

METHODS

Before starting with the field surveys, we inferred information on the distribution of acacias species from the national dataset "Flora Croatica Database" (http://hirc.botanic.hr/fcd). We found few records, all distributed along the coast, referring to the species *Acacia dealbata* Link, *A. retinoides* Schltdl. and *A. saligna* (Labill.) H. L. Wendl. f. No other records were found in the Croatian area by further inspecting the specimens stored in the Herbarium of the Faculty of Science, University of Zagreb (ZAHO). We also gathered general information on acacias growing preferences.

Following the above mentioned information, we decided to conduct field surveys along the Croatian coast. We carried on a first survey during the blooming period at the end of February 2014, inspecting north part of the Croatian coast (along about 480 km, roughly from Rovinj to Senj). We used the state roads along the coast (nr. 21, nr. 66, nr. 8) as a reference and the surrounding areas were checked for spotting acacias trees. We gave special attention to more urbanised areas, as the cities of Rovinj, Pula, Rijeka and Senj, where the main road was left aside to investigate private gardens inside the cities. During our field trip we have also visually checked for Acacia trees within natural vegetation, even though we mainly travelled along roads (it was easy to spot the trees because of their flowering). Locations of all the spotted plants were georeferenced by a global positioning system receiver. On each location we also recorded data as position of the plant (inside/outside gardens), state of flowering (as flowering phase), presence/absence of insects on flowers, identification of insects, inflorescence characteristics (number and diameter of flower heads). These data have then been combined with some ecological information, such as climatic parameters and soil reaction. We performed another field survey the second week of March 2014, inspecting the south part of the Croatian coast, from Cavtat up to Senj (about 550 km, along state road nr. 8). As for the first survey, the main road along the coast was used as a transect from which we made few departures in relation to urbanised areas, georeferencing any plant detected. We also visited the Botanical Garden on the Island of Lok-

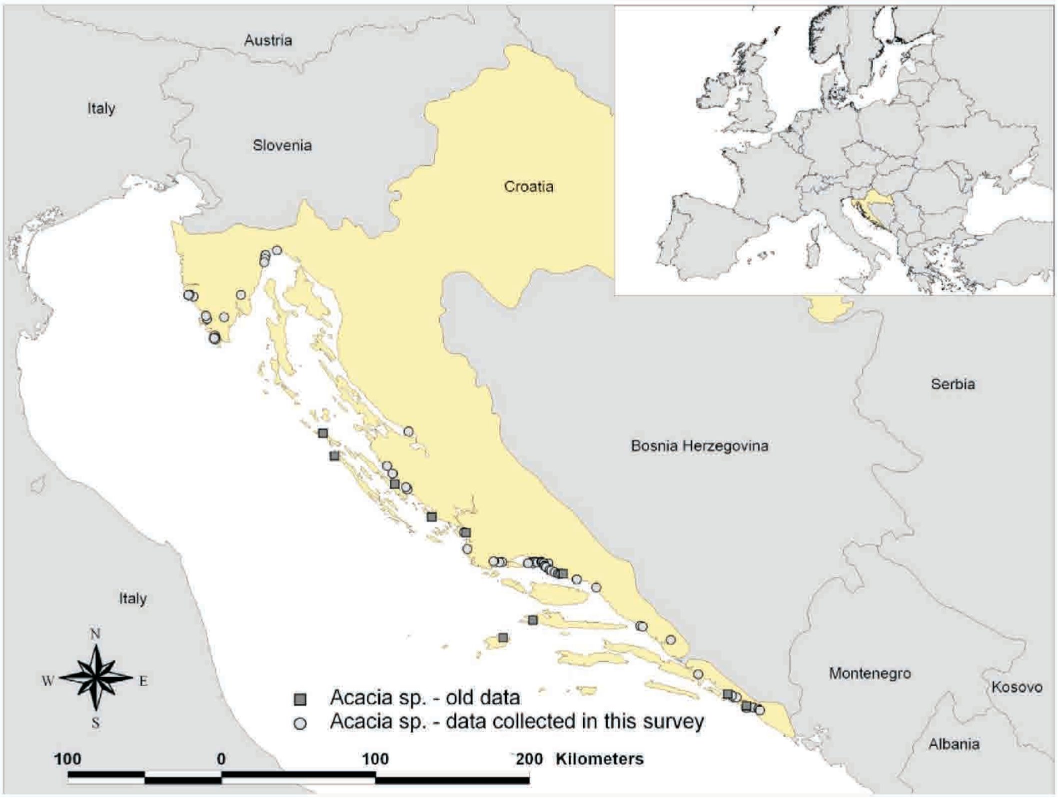

Figure 1. *Distribution of records for Acacia plants in Croatia. Dark grey squares refer to data of previously known localities obtained from Flora Croatica Database; light grey circles denotes localities that were achieved during this research (Inner map – location of Croatia in Europe).*

rum, founded in 1959 and containing many tropical and subtropical species from Australia and South America.

For what concerns biological data on *A. dealbata*, we analysed flowering phenology separately for data obtained on the two field surveys, since a gap of a week is expected to influence phenological state of observed plants. The phase of flowering was recorded as in Giuliani *et al. (6)*, on the base of a combination of flower head (FH) characteristics: presence/abundance, and percentage of FHs at different stages, i.e. bud/bloom/senescence, observed at the level of the whole crown. Accordingly we acknowledged six pheno-phases: Phen 1 (no or up to 30 % FH buds); Phen 2 (more than 30 % FH buds); Phen 3 (up to 50 % FH blooming); Phen 4 (more than 50 % FH blooming); Phen 5 (up to 30 % FH senescent); Phen 6 (more than 30 % FH senescent). Data on FH number per raceme were collected counting FHs on three racemes per plant (n = 3 trees of *A. dealbata* subsp. *dealbata* -ADD- and n = 5 trees of *A. dealbata* subsp. *subalpina* -ADS-). We recorded data on FH size by measuring the diameter of three central FHs on each of three racemes using an electronic calliper (Filotecnica Salmoiraghi Instruments; resolution: 0.01 mm; accuracy [<100 mm]: ± 0.02 mm), on 3 trees of ADD and 11 trees of ADS. We carried on observations on insect activity between 10:00 and 16:00 during sunny days.

Data about: elevation, slope, aspect (expressed as sin and cosine of angular values), mean and minimum yearly temperature and yearly precipitation were assigned to all the georeferenced localities. Using these environmental variables (with the exception of the elevation that is correlated with climate data) we have produced a map of suitable habitats by MaxEnt method *(27)*.

We collected soil samples to a depth of 15 cm after the removal of the leaf litter (if present), both near the acacias trees, and in the nearest location under natural or semi-natural vegetation, and left them to air-dry. Collection took place on few locations throughout the distribution area of *A. dealbata* from north to south, because it was not always possible to contact the owners of private gardens or to get the permission. We measured soil reaction with a WTW pH330i pH-meter on 10 g of soil in 25 ml of distilled H_20, as well as in 25 ml of potassium chloride (KCl), after 30 minutes with occasional stirring. We have used both methods to make easier comparability of acacia related soil pH obtained in other surveys.

RESULTS

Data collected during the first survey contributed with 40 locality records for *A. dealbata*, new to the Croatian database and the surveyed area. From the second survey,

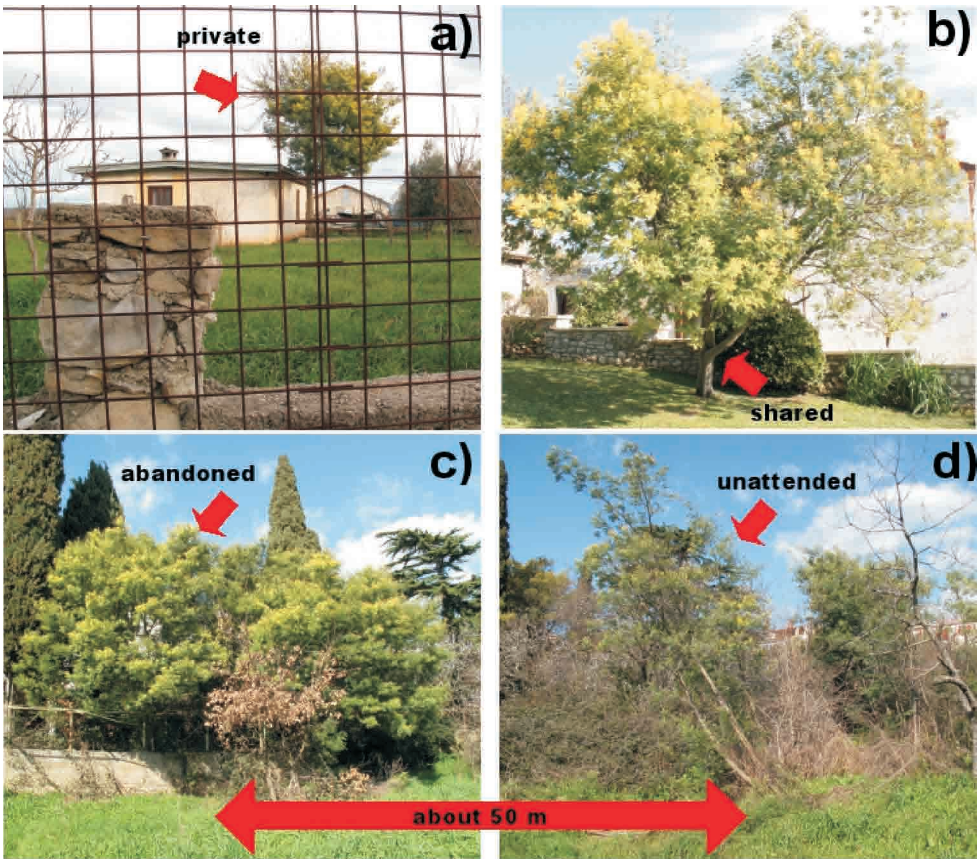

Figure 2. *Location of Acacia plants inside private (a) or shared (b) gardens. Natural spread of plants outside abandoned (c) or unattended gardens (d) have also been observed.*

other 107 new records for *A. dealbata* and 13 for other *Acacia* species were added. A final map with *Acacia* distribution has been produced (Figure 1).

In Croatia, *A. dealbata* plants are found exclusively in urbanised areas, their presence ascribable to their use as ornamentals in private and shared gardens (Figure 2 a,b). 86.8 % of all records were associated to private and 3.5 % to shared/public gardens. The remaining 9.7 % of records, anyway, refers to plants grown in abandoned or unattended gardens (Figure 2 c,d). Sprouts could be observed at short distance from trees, possibly resulting from vegetative reproduction; from few owners' interviews, it emerged that gardening activities reduce their presence and survival. In unattended gardens, many sprouts of considerable height were observed and the presence of other close trees could be attributed to seed regeneration. In the southern part of the coast, close to Dubrovnik, we also recorded some areas where acacias are commercially grown for flower market.

Since the survey took place during the flowering season, we recorded biological data as the state of flowering and the presence of insects acting as potential pollinators. During the first survey (Figure 3, upper part), the first flowering phases where missing, while four phenological phases were recorded: Phen 3, Phen 4, Phen 5 and Phen 6. During the second survey, the same four phenological phases were recorded, but with a dominance of the last one, clearly indicating that by the beginning of March *A. dealbata* flowering was over (Figure 3, lower part). Very few records of insects visiting the plants have been documented; 55.6 % of all insect observations were recorded at Phen 3 and 4, therefore associated with the full bloom of the plant. Nonetheless, also senescent flowers attracted some insects. Insects observed belonged to the families Hymenoptera (the more frequent being *Bombus terrestris* and *Apis mellifera*), Diptera (Syrphidae), Coleoptera (Cocinellidae) and Hemiptera.

We detected morphological differences among *A. dealbata* plants (especially leave size, see Figure 4, and flower head size, see Table 1). These differences, following a detailed bibliographic research *(28)*, may be ascribed to the presence of two subspecies: *Acacia dealbata* Link. subsp. *dealbata* (ADD) and *Acacia dealbata* Link. subsp. *subalpina* (ADS). On a total of 135 ascertained trees, 95.6 % were identified as belonging to ADS. Data on FH number per raceme and FH size were compared between the two subspecies (Table 1). ADD showed on average a statisti-

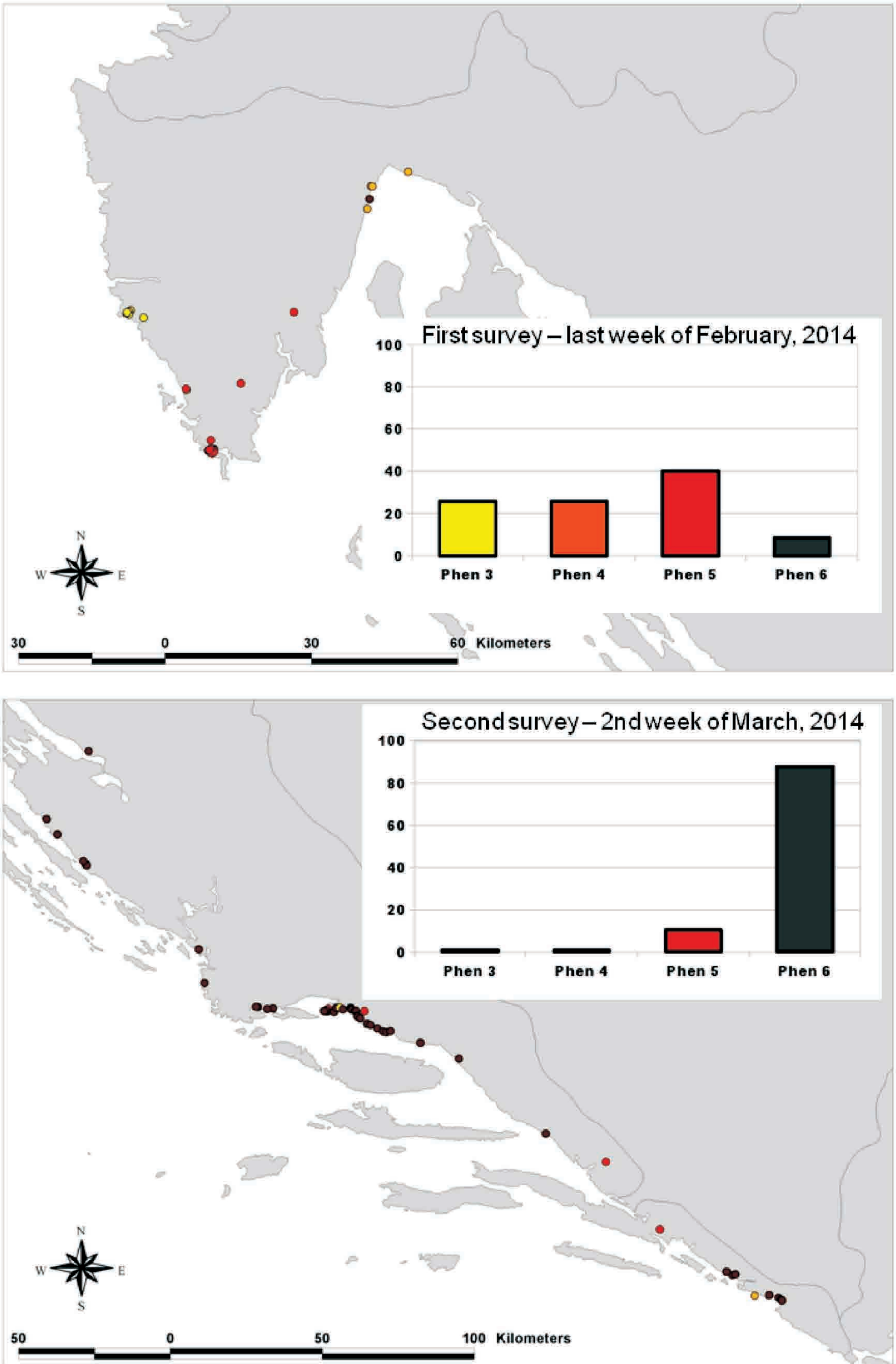

Figure 3. *Percentage of plants in a given phenological phase (graph embedded in each map) and their distribution, observed during the two field surveys.*

TABLE 1

Differences on no. of flower heads and their size between the two subspecies of *Acacia dealbata* recorded along the coast of Croatia.

	A. dealbata dealbata	A. dealbata subalpina
Average no. FH/raceme (±SE)	18.78 ± 1.43	12.73 ± 1.11
	T test, p = 0.003	
Average size of FH (mm, ±SE)	8.60 ± 0.14	7.86 ± 0.15
	T test, p = 0.002	

Figure 4. *Morphological differences between leaves of the two acacias subspecies recorded: on the left, Acacia dealbata subsp. subalpina; on the right Acacia dealbata subsp. dealbata.*

cally significant higher number of FH/raceme (T test, t = 3.327, df = 22, p = 0.003) than ADS. Similarly, the average size of FH was significantly larger in ADD than ADS (T test, t = 3.298, df = 70, p = 0.002).

The visit to Lokrum Botanical Garden allowed the investigation of an experimental area established for acclimatisation of alien species from Australia and South America. A specific area is especially dedicated to *Acacia* species and 18 species were supposed to have been planted in the garden, but unfortunately tags are missing for the majority of them. One third of the species named in an original list could not be associated to any currently valid scientific name and need further investigations. Other four species could not be ascertained, and the remaining eight were carefully controlled and their location in the garden finally linked with their accepted scientific name.

MaxEnt analysis for *A. dealbata* yielded a map of potential habitats (Figure 5) that was congruent with the majority of our field data, with the exception of the eastern part of Istria. Indeed this area was not recognised as a suitable habitat by the MaxEnt model, while we recorded several acacia plants (see Figure 1). Contrary to that, large areas north of Šibenik and Zadar (in the middle of the coastal region) were supposed to be suitable for acacias growth, but our field observation did not support this. The climatic parameters giving the highest contribution to the MaxEnt model were the minimum and mean yearly temperature (47 % and 37.2 % respectively), followed by northness and precipitation (7.5 % and 7 %

TABLE 2

Soil reaction measured on soil samples collected at few localities where acacias trees were observed. Samples were collected under acacias trees and at close natural or semi-natural sites. Coordinates (x;y) corresponds to Bessel ellipsoid, Gauss-Krüger projection (Transverse Mercator), 5th zone.

	Sites with *A. dealbata*		Sites without *A. dealbata*	
Location (x;y)	pH(H_2O)	pH(KCl)	pH(H_2O)	pH(KCl)
5411295; 4964292	7.14	6.34		
5411327; 4964268	5.90	5.12	6.22	5.44
5410724; 4964969	7.48	6.92	7.84	7.04
5410600; 4964850	7.25	6.63		
5548200; 4852700	7.66	7.18	8.07	7.42
5552169; 4849242	7.86	7.23	8.08	7.22
5615300; 4819250	8.17	7.3	7.96	7.29
5756320; 4724950	6.23	5.28	7.65	6.99

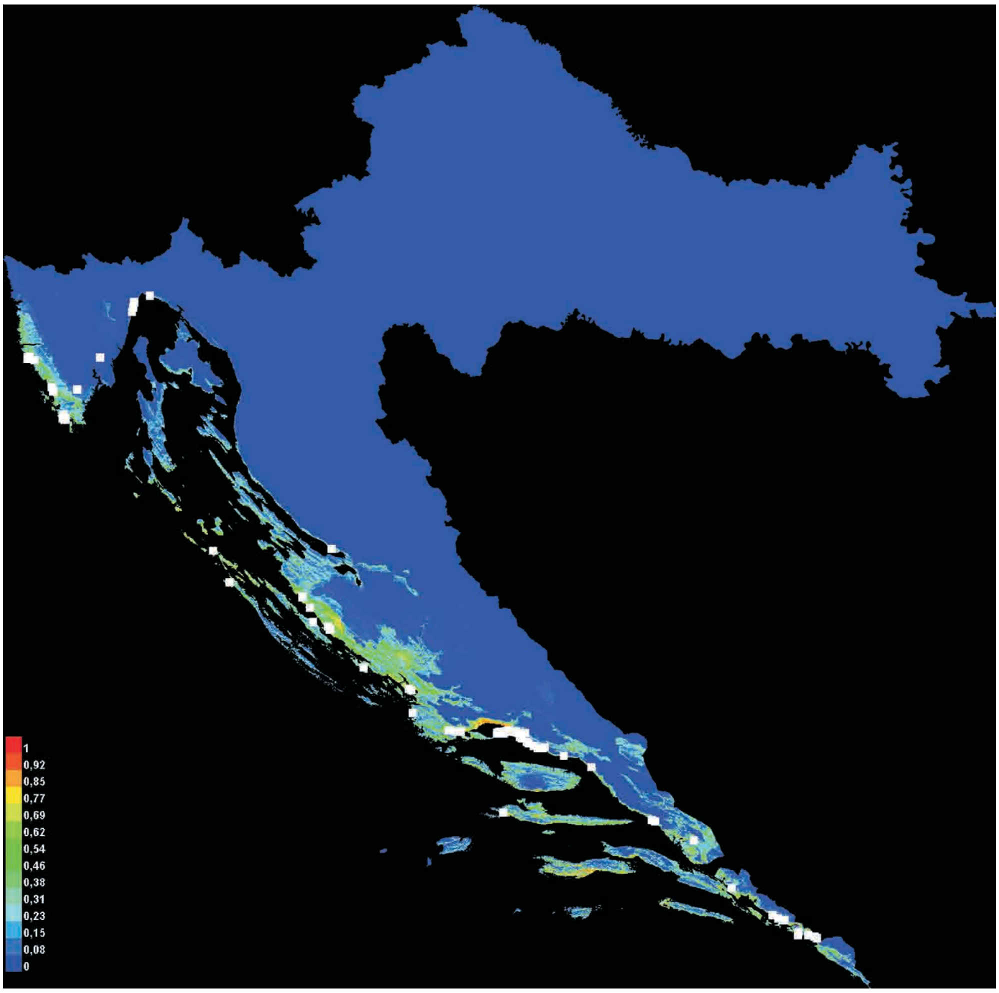

Figure 5. *MaxEnt map of suitable habitats in Croatia for Acacia dealbata Link based on 98 presence data (white squares) and six environmental variables (minimum and mean temperature, precipitation, slope, sin and cosin of aspect). Number (0 – 1) express results of logit function, where 0 denotes 0% of probability for habitat suitability i.e. potential for Acaccia occurrence and 1 denotes 100% of it.*

respectively); higher temperatures and more southern aspects contributed positively to suitability for acacias.

Acacias distribution is limited to the warmest areas, with as much as 90 % of all localities situated at elevations lower then 80 m. a.s.l., having absolute minimum yearly temperature above 1.1 °C, and mean temperature above 15.1 °C. Yearly precipitation ranged from 820 to 1200 mm (10 % and 90 % percentile). Analyses of soil acidity (Table 2) did not show unequal relation of acacia sites and nearby acacia-free samples, although acacia positive sites generally tend to have lower pH values then their negative counterparts. In general, the majority of soil samples had neutral to slightly basic reaction.

DISCUSSION

Terrestrial plants are the most represented environmental taxon group among alien species. Tree invasions may bring forth important conservation problems, such as variation in landscape structures, reduced regeneration rates of native species, interferences on native plant-animal interactions. One of the most invasive tree genera in the world is *Acacia*, due to the large influence of human-driven activities, to its ability in adapting to new environments and thanks to its massive vegetative regeneration and to changes brought to soil characteristics *(16-19)*.

In Croatia few data existed on this genus before this work, our surveys highlighted that acacias are far more abundant than previously thought. As expected, the Adriatic coast offers a suitable climatic environment for acacias growth and the huge amount of records provided by the survey confirmed it. The abundance is especially connected to the use of *A. dealbata* as ornamental in private and shared gardens; unlike in Mediterranean countries where it is clearly invasive, in Croatia *A. dealbata* was never widely used in forestry. Some sources report a large number of ”mimosa” trees, grown for commercial purposes further south in Montenegro, but without information on their invasion status *(29,* http://www.montenegro.com/city/djenovici*)*. The introduction of exotic species through gardening trade has been recognised as an actual gap in the European legislation that should be urgently addressed *(9, 30)*. In several cases we observed that an active gardening activity may control the vegetative sprouts, contrasting the natural potential to expand that the species revealed in unattended environments. Anyway, no plants have been recorded in natural habitats; other restrictions to acacias growth are probably involved in its actual distribution.

An additional reflection should regard the exact taxonomic identification of the species: we found that in the majority of cases *A. dealbata* subsp. *subalpina* was grown. Even if data collection was not exhaustive (many plants were restricted to private non-accessible gardens), the preliminary information highlighted differences in the biological characteristics: FH/raceme was generally lower than expected from literature *(28,* data from the native home range of the species*)*, which reported 22-42 FH/raceme for ADD and 13-34 FH/raceme for ADS, while we found values in the range 14-28, and 5-20, respectively. This may be an effect of the ecological conditions in the introduced range, while a subspecies may be preferred by gardeners due to higher resistance and better survival, which should be checked by thorough survey in the future.

About flower visitors, in this period of the year (late winter – early spring) it seems not surprising that few insects were recorded on *Acacia* inflorescences. Bees are the main pollinators of acacias in Australia *(31, 32)* and were recorded also in this study. Other insects were recorded: as already pointed out by Giuliani *et al. (6)*, the conspicuous flowering of *A. dealbata* at the end of the winter, often occurring before the flowering of the majority of native Mediterranean plants, may attract opportunist insects. Furthermore, for successful fertilization, the flowering phenology of acacias should match the activity period of pollinators, but this may not occur due to different reasons. Climatic variations from yearly averages, as was the case of the winter 2013/14 characterized as warm and extremely warm *(33-35)*, could have influenced bee and plant phenology, explaining the low number of insects recorded during these surveys.

Results from soil samples collected in this survey can be treated only as indicative, because of their limited number. However, under acacias we measured pH (H_20) values in a range from 7.14 to 8.17 (except one location with measured pH of 5.90): these values are much higher than those reported by Rodríguez-Echeverría *et al. (36)* and Lazzaro *et al. (16)*, where average pH values were not higher than 5.50. It seems that acacias tend to change soil reaction from acid in e.g Portugal and Spain towards less acid, while in Croatia it changes to less alcalic. Another potential environmental filter for spread of acacias in the eastern Adriatic could be attributed to water availability. Here, thin layer of litosol or cambisol is prevailing, which in combination with water permeable limestone bedrock may not provide favourable conditions: in fact, de Neergaard *et al. (37)* indicated *A. dealbata* as a water-demanding plant. It is possible that gardeners provide to young plants more favourable soil conditions (i.e. less alcalic than under the influence of naturally dominant carbonate parent rock), and more water in soil in early phases of growth, when plants are more sensitive. Without such help, acacias spread may be limited in natural conditions.

To conclude, notwithstanding acacias turned out to be very abundant along the Adriatic coast of Croatia, they can not be considered aggressive invaders as in similar climatic conditions in other Mediterranean countries. There are different factors that may turn out to have played a role in the current distribution of acacias in Croatia: the lack of use in forestation, fertilization success and development of viable seeds, soil reaction influence on seed germination and seedling growth. These several factors should be further investigated in the future. Finally, at the moment we can state that Australian acacias in Croatia seem to persist only in cultivation, therefore their current status may be denoted as alien – in cultivation *(38)*, with a local trend of becoming casual.

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