

Improving the early detection of alien wood-boring beetles in ports and surrounding forests

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Summary

1. International ports are generally considered the most likely points of entry for alien wood-boring beetles. A better understanding of the factors affecting their arrival and establishment at ports and their surrounding areas is of utmost importance to improve the efficacy and the cost-effectiveness of early detection programmes. Our work aimed at understanding how port size and the characteristics of the landscape surrounding the port, in terms of forest cover and forest composition, influence the occurrence of alien wood-boring beetles.
2. From May to September 2012, 15 Italian international ports and the surrounding forests were monitored with multi-funnel traps baited with a multi-lure blend (α -pinene, ethanol, ipsdienol, ipsenol, methyl-butenol), three in each port and three in forests located 3–5 km away from the port. We identified both alien and native Scolytinae, Cerambycidae and Buprestidae beetles.
3. Fourteen alien species, among which four are new to Italy, were trapped. Alien species richness was positively related to the amount of imported commodities at the port scale. Broadleaf forests surrounding ports received larger number of alien species than conifer forests. By contrast, total forest cover in the landscape surrounding ports was positively related to the occurrence of native but not alien species. The alien and native species richness was higher in the surrounding forests than in the ports.
4. *Synthesis and applications.* The simultaneous use of traps in ports with large volume of imported commodities and in their surrounding broadleaf forests can strongly increase the probability of alien wood-boring beetle interceptions. The identification of sites where the arrival and establishment of alien species is more probable, combined with an efficient trapping protocol, can substantially improve the efficacy of early detection. Similar approaches may be used in other countries as early warning systems to implement timely measures to eradicate or contain alien invasions at the European scale.

Key-words: bark beetles, exotic species, forest pests, invasion, jewel beetles, landscape, long-horn beetles, monitoring, species interception, surveillance

Introduction

Wood-boring beetles (Insecta, Coleoptera) are among the most important tree pests causing significant economic damage to forests world-wide (Brockhoff *et al.* 2006a). These insects are easily transported in almost all types of woody material, where they can easily hide from detection (Brockhoff *et al.* 2006b) and they are recognized as highly successful invasive group of species (Haack 2006;

McCullough *et al.* 2006). Every year, new alien species are intercepted and recorded as established in both Europe and North America (Work *et al.* 2005; Kirkendall & Faccoli 2010). This trend is expected to continue given the increase of international trade which will intensify colonization pressure (Levine & D'Antonio 2003; Hulme 2009; Kenis *et al.* 2009; Marini *et al.* 2011). Moreover, climate warming may reduce the thermal limitations now hampering the establishment of species coming from tropical or subtropical regions (Roques 2010). In this context, the early detection of alien species is of primary importance to improve the chance of effective eradication and

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provide better estimates of their arrival rates (Brockerhoff *et al.* 2006a; Rassati *et al.* 2014).

Although alien wood-boring beetles may arrive through fresh timber and plants for planting (Siitonen 2000; Piel *et al.* 2008; Liebhold *et al.* 2012), inspection data from USA and New Zealand indicate that crating, dunnage and pallets are the most common materials associated with these alien species (Haack 2001; Stanaway *et al.* 2001; Brockerhoff *et al.* 2006b). Pathway analyses indicated that international maritime ports are the most likely points of entry for alien wood-boring beetles because they receive large amounts of commodities that are commonly associated with wood packaging (Haack 2001; Brockerhoff *et al.* 2006a). In recent years, international cargo has been increasingly shipped in large containers that are difficult to inspect (Stanaway *et al.* 2001; McCullough *et al.* 2006). For this reason, several countries such as Australia, Canada, New Zealand and the USA combine traditional direct inspection of imported commodities with trapping programmes (Haack 2001; Brockerhoff *et al.* 2006a; Rabaglia *et al.* 2008; Wylie, Griffiths & King 2008; Rassati *et al.* 2014). Identifying the sites where the arrival and establishment of alien species is more probable is of utmost importance to improve the efficacy of early detection.

At the continental scale, the number of intercepted wood-boring beetles is often related to the volume of imported goods per state or country (Haack 2001; Huang *et al.* 2012). It is still unclear, however, if such a relation occurs also at the port scale. In addition, a few studies have investigated how the landscape surrounding high-risk sites can influence the establishment of alien species (Bashford 2008; Rabaglia *et al.* 2008). The surrounding landscape often consists of mosaics of urban areas, green spaces, crop fields or different types of forest where alien species are challenged by environmental and demographic stochastic forces that must be overcome to establish and spread (NRC 2002). For instance, there has been much debate on the effect of native tree density, diversity and distribution on the invasion success of wood-boring beetles (Brockerhoff, Liebhold & Jactel 2006; Colunga-Garcia *et al.* 2010). On the one hand, high tree species diversity in urban areas located around high-risk sites seems to be favourable for the establishment of alien species, as it can provide wide range of adequate hosts (Koch *et al.* 2011). On the other hand, urban areas with limited forest cover may hamper the invasion process as it cannot support many new insect establishments due to host limitation (Novak 1994; Koch *et al.* 2011). Moreover, an important role can be played by the composition and structure of forests located in these areas (Brockerhoff, Liebhold & Jactel 2006). Considering that most alien wood-boring beetles are polyphagous feeders on broadleaf hosts (Kirkendall & Faccoli 2010; Marini *et al.* 2011), the presence of mixed broadleaf rather than conifer-dominated forests can facilitate the establishment of these species. Understanding the role of these factors is important in

identifying which sites deserve special attention in surveillance programmes.

The main purpose of this study is to gain insights into the factors affecting the occurrence of alien wood-boring beetles in and around maritime ports. In particular, we investigated the relationships between the occurrence of alien wood-boring beetles and (i) the annual volume of imported commodities, (ii) the characteristics of the landscape surrounding each port in terms of forest cover and composition. First, as the amount of wood packaging materials is positively related to the amount of imported goods, we expected higher alien species richness in ports importing a high volume of commodities. Secondly, as most alien wood-boring beetles are polyphagous feeders on broadleaf hosts (Kirkendall & Faccoli 2010; Marini *et al.* 2011), we expected broadleaf forests to be more suitable to alien species invasions than conifer forests. Specifically, we will test whether it is sufficient to monitor only port areas or whether trapping should include both ports and surrounding forests. The analyses of the environmental factors affecting alien species occurrence will help identify the sites where the monitoring efforts should be concentrated to improve early detection effectiveness.

Materials and methods

SITE SELECTION AND EXPERIMENTAL DESIGN

Fifteen international ports located along the Italian peninsula and main islands (Sardinia and Sicily) were selected (Fig. 1, see Table S1, Supporting information). The ports covered a wide latitudinal gradient (min. Catania: 37°29'47"; max. Porto Nogaro: 45°47'49") and were selected considering three statistically orthogonal factors: volume of solid commodities imported per

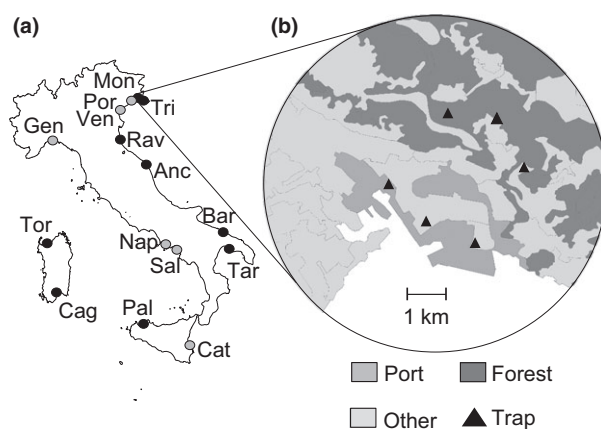


Fig. 1. (a) Geographical distribution of the 15 Italian surveyed ports and (b) an example of how the traps were set-up in each site. Black circles on the map indicate ports surrounded by conifer forest and grey circles indicate ports surrounded by broadleaf forest. Port name abbreviations: Ancona (Anc), Bari (Bar), Cagliari (Cag), Catania (Cat), Genova (Gen), Monfalcone (Mon), Napoli (Nap), Palermo (Pal), Porto Nogaro (Por), Porto Torres (Tor), Ravenna (Rav), Salerno (Sal), Taranto (Tar), Trieste (Tri), Venezia (Ven).

year, proportion of forest cover in the 10 km surrounding each port and forest composition (conifer- vs. broadleaf-dominated forest) (see Table S1, Supporting information). Regarding the first two factors, we selected ports in order to provide the widest possible range of values. Due to the low number of available ports at the national scale, it was not possible to have a balanced experimental design between conifer ($n = 9$) and broadleaf ($n = 6$) forests. In each of the 15 selected sites, we sampled insects inside the port and in a surrounding forest. The distance between the sampling points inside and outside the port was between 3 and 5 km.

The amount of imported commodities in 2011 was obtained from Assoporti (2012) or directly from the port authorities (Cagliari, Catania, Porto Torres and Trieste). We considered only the total volume of solid commodities because they are usually associated with wood packaging materials, while we excluded data about imported fluids such as oil or liquefied gas because they are not associated with wood packaging. Forest cover in the landscape surrounding each port was calculated in ARCGIS 10.0 (ESRI, Redlands, CA, USA) based on detailed digital aerial photographs (Google Earth). First, we digitized polygons representing both the port and the forest areas in Google Earth, excluding single trees or hedgerows with an area smaller than 100 m². Then using ARCGIS, we determined the centroid of each port and we established a 10-km radius buffer zone. Lastly, we calculated the percentage of landscape covered by forest after having excluded the sea area from the buffer zone. Each forest site selected as a survey point was classified visually by local forest health inspectors involved in the project into broadleaf or conifer forest according to the tree composition. The forest sites were representative of the dominant forest type occurring in the 10-km radius buffer around each port. Sites were assigned to one of the two forest categories when the dominant cover type was 80% or more. Conifer forests were mainly composed of pines, either Austrian *Pinus nigra* Arnold or Mediterranean *Pinus pinaster* Aiton and *Pinus pinea* Linnaeus pines, while broadleaf forests were always mixed stands composed mainly of oak *Quercus* spp., hophornbeam *Ostrya carpinifolia* Scop. and ash *Fraxinus* spp. species.

TRAPPING DESIGN AND LURES

In each of the 15 sites, six 12-unit black multiple-funnel traps (Econex, Murcia, Spain) were set-up, three inside the port and three in a forest site close to the port. The traps within the same environment (either port or forest) were at least 30 m apart. In Catania, only two traps were placed inside and outside the port. At each site, the traps were hung at about 2 m above the ground.

All traps were baited with a generic multi-lure blend composed of (–) α -pinene (Ultra High Release, release rate of 2 g day⁻¹; 90 days field-life at 20 °C), ipsenol (+50/–50; release rate of 0.4 mg day⁻¹; 90 days field-life at 20 °C), ipsdienol (release rate 0.4 mg day⁻¹; 90 days field-life at 20 °C), 2-methyl-3-buten-2-ol (release rate of 11 mg day⁻¹; 90 days field-life at 20 °C) and ethanol (release rate of 0.3 mg day⁻¹; 90 days field-life at 25 °C) provided by Contech Enterprises Inc. (Victoria, BC, Canada). These lures had been tested earlier and attract a wide variety of wood-boring beetles (Rassati *et al.* 2014). We did not add any liquid to the collection cups, and therefore used an insecticide (FERAG IDTM; SEDQ, Spain) to quickly kill the insects. We changed the lures after 3 months based on their expected field-life.

Trapping occurred from early May to late September 2012 (150 days). The number of trap checks varied from two to nine (average $n = 6.7$) in relation to restrictions to port access (see Table S1, Supporting information). The number of trap checks was not correlated with neither volume of import ($r_s = 0.22$, $P = 0.43$) nor forest cover in the landscape ($r_s = 0.43$, $P = 0.1$). Adults of the target groups (Scolytinae, Cerambycidae and Buprestidae) were stored in alcohol. Most individuals were identified on the basis of morphological features, but in a few cases, we used molecular techniques. In particular, DNA extraction was carried out following a salting out protocol based on the differential solubility of proteins and DNA at high salt concentrations (Patwary *et al.* 1994). The barcode region of the mitochondrial gene cytochrome oxidase I was amplified using universal primers (Folmer *et al.* 1994), and the obtained sequences were compared with those already deposited in the BoldSystem database (Ratnasingham & Hebert 2007). Species were classified either as native or alien according to the available literature (Wood & Bright 1992; Curletti 1994; Bense 1995; Pfeiffer 1995). We considered as alien all those species that are not native to Italy. This category can include species that are already established, previously intercepted but not yet established, or never intercepted before. We decided to consider as alien also those species already established in Italy for three main reasons: as the general aim of our study is to identify priorities for identifying the sites with the highest risk of invasion, the abundance and diversity of the established alien taxa can provide a clear indication of the suitability of a site to be invaded; secondly, every year new individuals of established species can arrive at different points of entry where they are not present yet, establishing a new population and starting a new invasion; thirdly, new individuals can increase the genetic diversity of the established population, improve their fitness and modify the possible impacts on native ecosystems.

DATA ANALYSIS

To account for the differences in trapping frequency and the variability due to the longer intervals between less-frequent trap checks, we used a generalized linear mixed-effects model to evaluate the effect of the time between trap checks on the mean number of species or abundance per trap. Then, we calculated the model residuals, and we used them as a response variable to test the effect of import volume (continuous variable), forest cover in the landscape (continuous variable), composition of forest site close to the port (categorical variable: conifer vs. broadleaf) and trap position (categorical variable: port vs. forest). The new response variable did not depend on the duration of the trapping. The model included the sampling site as a random factor to account for the spatial dependence of the sampling. The model was fitted using the ‘lme’ function in the package nlme for R version 2.15.1 (R Development Core Team 2012).

To compare the response of alien vs. native species, we used the same approach described above testing the effect of forest cover in the landscape, trap position and forest composition on native species richness and abundance. We did not include in the model the volume of imports as we did not expect any effect of this variable on native species.

Due to the relatively low number of replicates and the relatively high number of potential predictors, we used multi-model inference within an information-theoretic framework to

evaluate the role of the selected variables in explaining species richness and abundance (Burnham & Anderson 2002). Our information-theoretic approach compared the fit of all the possible candidate models obtained by the combination of our predictors using second-order Akaike's information criterion (AICc) corrected for small samples. The AICc is a measure of relative model fit, proportional to the likelihood of the model and the number of parameters used to generate it. The best-fitting model is the one with the lowest AICc. In a set of n models, each model i can be ranked using its difference in AICc score with the best-fitting model ($\Delta\text{AICc}_i = \text{AICc}_i - \text{AICc}_{\text{MIN}}$). The difference in AICc values indicates the relative support for the different models. A model is usually considered plausible if its ΔAICc is below 2 (Burnham & Anderson 2002). From the set of plausible models, we omitted the models with uninformative parameters, that is models with ΔAICc below two but including only one additional parameter compared to the best model (Arnold 2010). For each model i , we also calculated an Akaike weight (w_i), which is the probability that model i would be selected as the best-fitting model if the data were collected again under identical circumstances (Burnham & Anderson 2002). To gauge the relative importance of each predictor, we summed the w_i across the models in the set ($\sum w_i$) in which the predictor occurred. The multi-model inference analyses were performed using the MuMIn package for R (Barton 2010).

To test the similarity between the species recorded in each port and the surrounding forests, we used the Simpson's Similarity Index (Magurran & McGill 2010). A value close to 1 indicates that the two communities are very similar in the two environments. For each pair of port and surrounding forest, we computed the Simpson's Similarity Index for natives and aliens, separately. We used a one-way ANOVA to test difference in similarity between alien and native species.

To describe the influence of our selected factors on species composition, ordination methods were applied. The response variable was the species by site matrix based on species presence/absence. A preliminary detrended correspondence analysis (DCA) was performed. The largest DCA gradient length, expressed in standard deviation (SD) units of species turnover, of the first four DCA axes was below 3 SD units. Thus, the use of linear-based ordination models was appropriate for these data (ter Braak & Šmilauer 2002). A principal component analysis (PCA) was performed to extract the main part of the variability related to species composition. The factors trap position and forest composition were superimposed on the ordination plot to describe the similarity in species composition between ports and surrounding forests.

Results

GENERAL RESULTS

Overall, we collected 81 species of wood-boring beetles (see Table S2, Supporting information). Scolytinae represented the most abundant and diverse group with 49 species and 40 374 individuals, followed by Cerambycidae (26 species and 1371 individuals) and Buprestidae (six species and eight individuals). Sixty-seven species were native (82.7%) and 14 (17.3%) were alien species, including 11 Scolytinae and three Cerambycidae. Among alien species,

six Scolytinae [*Ambrosiodmus rubricollis* (Eichhoff), *Cyrtogenius luteus* (Blandford), *Gnathotrichus materiarius* (Fitch), *Hypothenemus eruditus* Westwood, *Xylosandrus crassiusculus* (Motschulsky), *Xylosandrus germanus* (Blandford)] and two Cerambycidae (*Phoracantha recurva* Newman, *Xylotrechus stebbingi* Gahan) were already known to be established in Italy. The other six species were recorded for the first time in Italy [the Scolytinae *Liparthrum colchicum* Semenov, *Pseudothammurgus scrutator* (Pandellè), *Xyleborus ferrugineus* (Fabricius) and *Xyleborus volvulus* (Fabricius)] or previously collected in earlier surveys [the scolytid *Ernoporicus caucasicus* (Lindemann) and the cerambycid *Cordylomera spinicornis* (Fabricius)] (Cola 1971) but were not considered to be established in Italy (see Table S2, Supporting information). No alien Buprestidae were trapped.

Of the eight alien species considered as established, one species was trapped exclusively in ports, two were trapped exclusively in the surrounding forests and five were found in both environments (see Table S2, Supporting information). Regarding the six species not considered as established, three were trapped only in ports and three only in the surrounding forests, while no species were trapped in both environments. Among the alien wood-boring beetles trapped exclusively in ports, two were species associated only with conifers and two only with broadleaf trees, while 75% of the species trapped exclusively in the surrounding forests were associated with broadleaf trees (see Table S2, Supporting information). Considering the alien species trapped in both environments, three were species associated only with broadleaf trees, one only with conifers and one with both conifer and broadleaf trees. The Scolytinae *C. luteus* and *X. germanus* were the most commonly collected species, with 220 and 104 individuals, respectively. By contrast, four species were represented by only one individual each, namely the Scolytinae *A. rubricollis* and *G. materiarius* and the Cerambycidae *C. spinicornis* and *P. recurva*.

Of the 67 native species, 10 were trapped exclusively in ports, 23 were trapped exclusively in the surrounding forests and 34 in both environments (see Table S2, Supporting information). The bulk of the species trapped exclusively in ports was associated only with conifers (60%), while the others were associated only with broadleaf trees (20%) or both broadleaf and conifer trees (20%). The opposite trend was observed considering the species trapped exclusively in the surrounding forests given that most were represented by species associated only with broadleaf trees (43%). Amongst the species trapped in both environments, the main part was associated with conifers (58%), while only 29% were associated exclusively with broadleaf trees. *Orthotomicus erosus* (Wollaston) and *Hylurgus micklitzi* Wachtl were the two most commonly collected Scolytinae, with 24 801 and 10 829 individuals, respectively, while *Spondylis buprestoides* (Linnaeus) was the most commonly collected cerambycid, with 840 individuals.

SPECIES COMPOSITION

The Simpson's Similarity Index for alien species between the port and the surrounding forests (0.1) was significantly lower than that for native species (0.69) ($P < 0.001$, $n = 15$).

For alien species, the first two principal components explained 33.4% and 14.8% of the total variation in species composition. The first axis indicated a clear separation between ports and surrounding forests, while the second axis was not associated with either trap position or forest composition (Fig. 2). Several ports located at very different latitudes presented similar alien species composition. For native species, the first two principal components

explained 18.7% and 11.1% of the total variation in species composition. The first axis mostly separated sites according to latitude. Contrary to the results of alien species, several ports presented very similar native species composition with the corresponding surrounding forests.

DRIVERS OF SPECIES RICHNESS AND ABUNDANCE

We found three plausible models explaining the occurrence of total alien species in ports and surrounding forests (Table 1a). The model weights were generally high, indicating low model selection uncertainty. The sum of model weights of the variables indicated that the species richness was influenced mainly by trap position

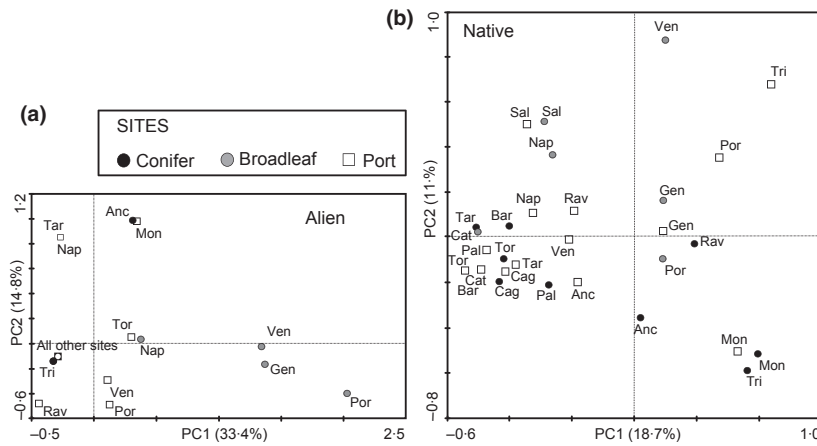


Fig. 2. Principal Component Analysis (PCA) plot for (a) alien and (b) native wood-boring beetles in ports and surrounding forests. Abbreviations of the sites are according to Fig. 1 and Table S1 (Supporting information).

Table 1. Plausible candidate models (within 2 ΔAICc of the top model) explaining species richness and abundance of wood-boring beetles (Scolytinae, Cerambycidae, Buprestidae) trapped during the survey, separately for alien and native species. Models are ranked according to their second-order Akaike's information criterion (AICc). Parameter estimates and model weight (w_i) are reported. For each tested variable, $\sum w_i$ indicates the sum of model weight

	Aliens					Natives			
	Best model	2nd	3rd	4th	$\sum w_i$	Best model	2nd	3rd	$\sum w_i$
a) Species richness									
ΔAICc	0	1.33	1.82	–	–	0	0.13	1.19	–
Model weight	0.27	0.14	0.11	–	–	0.34	0.32	0.18	–
Intercept	3.2×10^{-01}	4.8×10^{-01}	3.9×10^{-01}	–	–	3.5×10^{-01}	-2.2×10^{-01}	7.9×10^{-01}	–
Import	0.02	–	0.02	–	0.64	n.a.	n.a.	n.a.	–
Forest cover	–	–	-0.005	–	0.26	0.03	0.03	–	0.69
Forest composition	*	*	*	–	0.80	*	–	*	0.44
Trap position	*	*	*	–	0.82	*	*	*	0.94
Forest composition × Trap position	*	*	*	–	0.57	–	–	–	–
b) Abundance									
ΔAICc	0	0.91	1.09	1.10	–	0	1.34	1.71	–
Model weight	0.24	0.15	0.14	0.14	–	0.41	0.21	0.17	–
Intercept	8.9×10^{-01}	6.8×10^{-01}	8.5×10^{-01}	1.1×10^{-01}	–	7.0×10^{-01}	4.4×10^{-01}	3.6×10^{-01}	–
Import	–	0.02	0.03	–	0.42	n.a.	n.a.	n.a.	–
Forest cover	–	–	-0.01	-0.01	0.39	–	0.02	–	0.36
Forest composition	*	*	*	*	0.86	–	–	*	0.32
Trap position	*	*	*	*	0.86	*	*	*	0.93
Forest composition × Trap position	*	*	*	*	0.68	–	–	–	–

*Indicates that the categorical variable was included in the model.
n.a. Not applicable: import volume was not tested for native species.

($\sum w_i = 0.82$), composition of the forests surrounding ports ($\sum w_i = 0.80$), volume of imported commodities (positively, $\sum w_i = 0.64$) and interaction between trap position and composition of the forests surrounding ports ($\sum w_i = 0.57$). Forest cover in the landscape was the least important variable. The mean number of alien species standardized by the time between trap checks was higher in surrounding forests (0.041 ± 0.012) than in ports (0.006 ± 0.002) when the forests were mainly composed by broadleaf species ($n = 6$), while these values were similar for conifer forests (Fig. 3a).

We also found four plausible models explaining the alien wood-boring beetle abundance (Table 1b). The sum of the model weights of the variables indicated that the latter was influenced mainly by trap position ($\sum w_i = 0.86$), composition of the forests surrounding ports ($\sum w_i = 0.86$), interaction between trap position and forest composition ($\sum w_i = 0.68$) and volume of imported commodities ($\sum w_i = 0.42$). Forest cover in the landscape was the least important variable. As for species richness, the mean number of alien individuals standardized by the time between trap checks was higher in broadleaf forests (0.19 ± 0.22) than both in conifer forests (0.017 ± 0.011) and ports (0.015 ± 0.007). Considering separately the richness and abundance of species intercepted but not yet established, we did not find any significant effect of the tested variables.

We found three plausible models explaining the native species richness (Table 1a). The model weights were generally high indicating low model selection uncertainty. The sum of model weights indicated that the native species richness was influenced by trap position ($\sum w_i = 0.94$) and positively by the forest cover in the landscape ($\sum w_i = 0.69$). The composition of the forests surrounding ports was the least important variable. The mean number of native species standardized by the time between trap checks was higher in conifer forests (0.25 ± 0.04) than

both in broadleaf forests (0.18 ± 0.04) and ports (0.15 ± 0.03) (Fig. 3b).

We also found three plausible models explaining the native wood-boring beetle abundance (Table 1b). The sum of model weights indicated that the latter was mainly influenced by trap position ($\sum w_i = 0.93$). The forest composition was the least important variable. The mean number of native individuals standardized by the time between trap checks was higher in conifer forests (24.7 ± 14.81) than both in broadleaf forests (6.33 ± 5.14) and ports (1.96 ± 0.47).

Discussion

Our nationwide study clearly identified where best to concentrate surveillance efforts to effectively intercept alien wood-boring beetles. In order to increase the probability of detecting alien species soon after their arrival, extensive monitoring programmes should be concentrated in ports with large volumes of imports and in the surrounding broadleaf forests, as the combination of these two conditions seems to provide the most favourable circumstances for trapping alien wood-boring beetles.

We found a significant effect of the volume of imported commodities on the alien species richness both in the ports and the surrounding forests indicating that import volume may have a key role in favouring alien invasions. Although a species can be already established, every year new individuals can arrive in the same points of entry or in areas where the species are not yet present, potentially establishing a new population and starting a new invasion. Previous studies have already shown the importance of several socio-economic indicators on alien species richness, but these analyses were mostly performed at the continental scale (Levine & D'Antonio 2003; Hlasny & Livingston 2008; Hulme 2009; Essl *et al.* 2011; Huang *et al.* 2012). For instance, Haack (2001) reported that the number of interceptions of alien wood-boring beetles in the US was positively correlated with the value of general imports per US state, but this relationship was not tested at smaller spatial scales. Wood packaging materials associated with imports are commonly found discarded at ports and such materials are often associated with alien wood-boring beetles (Haack 2001; Stanaway *et al.* 2001; Brockerhoff *et al.* 2006b). Wood packaging materials may transit through the ports to their final destination or, when broken and not usable, stored at the ports before being sent to companies authorized to recycle or destroy the wood. Therefore, there are many opportunities for adult insects to emerge from infested wood packaging and disperse in the surrounding habitats. In fact, despite ISPM-15 (IPPC 2013), some wood-boring beetles are apparently able to survive the approved treatments or colonize and develop in wood after them (Haack & Petrice 2009). Moreover, considering that some treatments may be improperly applied, either knowingly or because of faulty equipment or facilities (Haack & Petrice 2009), the

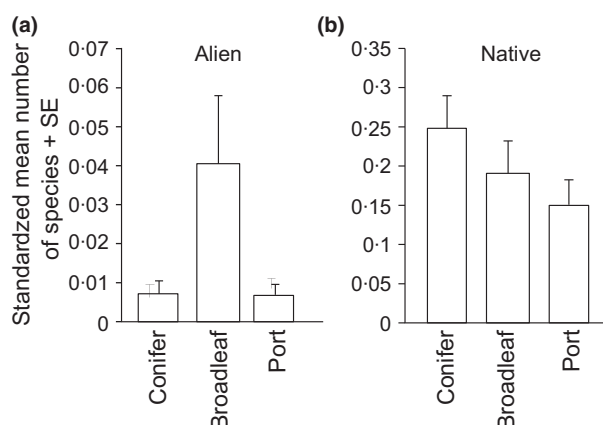


Fig. 3. Mean (\pm SE) number of alien and native species trapped per trap check in conifer forests, broadleaf forests and ports, standardized by the time between trap checks (number of species/duration of the sampling).

risk related to the movement of wood packaging materials is still relatively high. Our results suggested that resources for surveillance programmes should be spatially concentrated in ports that import high volumes of solid commodities.

We also found that alien wood-boring beetle richness and abundance in the forest surrounding ports were strongly influenced by the tree composition, with broadleaf-dominated forests supporting more alien species and individuals than conifer-dominated forests. As the majority of alien species are generalist insects feeding on several broadleaf genera (e.g. ambrosia beetles) (Kirkendall & Faccoli 2010; Marini *et al.* 2011), broadleaf forests surrounding ports represent suitable habitat for their establishment. These areas have been recognized as high-risk sites in previous studies as the wood-boring beetles arriving at points of entry can disperse to neighbouring areas exploiting their ability to fly and find suitable hosts and habitats (Bashford 2008; Rabaglia *et al.* 2008). However, this result was independent of the amount of forest cover in the landscape. Although it could be expected that an area with limited forests would be less suitable for the establishment of introduced insects (Novak 1994; Koch *et al.* 2011), our results show that alien wood-boring beetles can be present even in landscapes with very low amounts of forest area. Previous studies have suggested that urban areas, which are usually characterized by a wide range of native and alien tree species in parks, gardens and along streets, may provide adequate tree hosts for invaders that would not otherwise become successfully established (Bashford 2008; Koch *et al.* 2011). The role of plantings in the landscape surrounding high-risk sites is, however, still largely under-investigated (Reed & Muzika 2010). The movement of alien insects from the port to the surrounding areas is almost inevitable; therefore, these areas are recognized as a crucial point in determining the success or the failure of the invasion process (Bashford 2008; Rabaglia *et al.* 2008). By contrast, for native species, we found that the proportion of forest cover in the surrounding landscape was positively related to the species richness, independently of the forest composition.

Lastly, we found that alien and native species richness and abundance were influenced by trap position, that is they were higher in the surrounding forests than inside the ports. The presence of woody materials in ports is, in fact, not constant over time and space, with commodities periodically unloaded, shipped or moved (Stanaway *et al.* 2001). Forest areas provide a more stable habitat with a larger variety of potentially suitable hosts and most of the wood-boring beetles emerging from wood packaging materials are not expected to reproduce inside ports but instead to fly away searching for suitable hosts. Our results confirmed this trend, as the communities of alien wood-boring beetles trapped in ports were clearly different than the beetle communities trapped in the surrounding forests. For these reasons, setting traps in both environments will increase the chances of detecting alien

wood-boring beetles (Bashford 2008; Rabaglia *et al.* 2008; Wylie, Griffiths & King 2008). This trapping protocol, integrated with the traditional inspections carried out by plant health inspectors, can strongly increase the possibility of early detection of alien species. Other countries, such as New Zealand and USA, have already implemented similar approaches, with a number of intercepted alien species attesting to the importance of such trapping programmes (Brockhoff *et al.* 2006a; Rabaglia *et al.* 2008) and confirming their efficiency as an early warning system to trigger eradication or measures to contain the invasion. Regarding the native species, the results indicated that the wood-boring beetle communities trapped in ports were similar to those trapped in the surrounding forests, suggesting a potential exchange of species between the two environments. These individuals may fly from the forests towards the ports where they can colonize and complete development in wood packaging material (Haack *et al.* 2014), especially when bark is present (Haack & Petrice 2009). Traps can give information on the native species most commonly found in ports. These native species may constitute a pool of invaders that can be moved outside the country through international trade.

The development of early detection methods for alien species is a crucial step when implementing rapid response systems, effective eradication and suppression protocols for invasive pests (Pluess *et al.* 2012). If alien wood-boring and bark beetles are quickly detected, site-specific phytosanitary measures can be implemented and a timely action plan can be produced. However, due to the limited resources available for early detection, it is necessary to identify the most vulnerable locations where to concentrate surveillance efforts. Our countrywide survey of wood-boring beetles, with fourteen trapped alien species, among which four new to Italy, has provided clear indications of the most susceptible sites to invasions and has shown the efficacy of such a monitoring protocol. In particular, traps baited with attractive lures should be deployed both in ports and their surrounding forests, concentrating the efforts in ports with large volumes of imported commodities and in broadleaf forests surrounding them. Besides providing early detection of new invasions of alien pests, our trapping program can provide useful data on the geographical range of established alien species in support of regulatory controls or specific management programmes. Further benefits of our approach will be related to the potential reduction of the economic and environmental losses due to alien species. As traps and lures are fairly cheap and simple to use, pest management officials and the general public can be easily engaged in the fight against alien species (USDA-APHIS 2011). Significant challenges, however, still remain in the development of more effective surveillance tools for alien wood-boring beetles. In particular, more research efforts should be placed to develop more effective lure and trap systems for the large pool of potential alien wood-boring beetle species that may arrive in the future. At the same

time, the analyses of native species trapped in ports can provide useful information on the species that constitute a pool of invaders that can be moved outside the country through international trades. Similarly, investigations on host preferences of commonly intercepted alien species could be conducted in both their native range and in the new environment (in a quarantine facility) to learn which tree species are most at risk as well as to help prioritize where surveys should be conducted.

Acknowledgements

The authors thank the staff of the Regional Plant Protection Organization of Campania (Raffaele Griffo, Vincenzo Martino), Emilia Romagna (Gino Tallevi, Paolo Solmi), Friuli-Venezia Giulia (Iris Bernardinelli, Giancarlo Stasi, Carlo Frausin), Liguria (Matteo Benedetti), Marche (Emanuela Ricci), Puglia (Antonio Dangelico), Sicilia (Giuseppe Bono, Roberto Federico, Filadelfo Conti, Giuseppe Marano, Alfio Cutuli, Sebastiano Privitera), Sardegna (Wilson Ramassini) and Veneto (Marco Vettorazzo, Enrico Chiariot) for field assistance and collaboration during research; Mauro Simonato for the help with genetic analyses and Robert A. Haack for comments on an earlier draft of this paper. The authors are also grateful to two anonymous reviewers for the insightful comments that improved early drafts of the manuscript. This study was supported by the EU Seventh Research Framework Program (FP7) projects Q-DETECT (Development of detection methods for quarantine plant pests for use by plant health inspection services-Grant No. 245047).

Data accessibility

Alien and native species richness and abundance per site: data available from the Dryad Digital Repository: <http://doi.org/10.5061/dryad.r7j3s> (Rassati *et al.* 2015).

Parameters characterizing the 15 surveyed ports: uploaded as online supporting information.

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Received 29 April 2014; accepted 8 September 2014

Handling Editor: Yann Clough

Supporting Information

Additional Supporting Information may be found in the online version of this article.

Table S1. Parameters characterizing the 15 surveyed ports.

Table S2. Abundance and host category of the wood-boring beetles trapped during the survey.