Host Preference of *Tomicus piniperda* and *Tomicus destruens* for Three Pine Species

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Abstract – Host preference of four *Tomicus* portuguese populations was studied using maritime pine (*Pinus pinaster* Aiton), Aleppo pine (*Pinus halepensis* Miller) and stone pine (*Pinus pinea* L.). Beetle preference was evaluated using paired freshly cut logs of different species with similar diameter and bark characteristics. One of the tested populations was identified as *Tomicus piniperda* and the others as *Tomicus destruens*, suggesting that this species is dominant in Portugal. Maritime pine was preferred to Aleppo and stone pine. The comparisons between Aleppo pine and stone pine show that the two populations of *Tomicus* from South of Portugal preferred stone pine and the two populations from North Portugal preferred Aleppo pine.

I. Introduction

Tomicus piniperda L. and *Tomicus destruens* Wollaston are closely related species with similarities in their biology. The pine shoot beetle, *T. piniperda*, is an important pine pest widely distributed across Europe and Asia (Ye, 1991, Långström *et al.* 1992,), while *T. destruens* has a more restricted distribution, infesting pines in the Mediterranean region (Carle, 1974, Mendel *et al.*, 1985). Nevertheless, sympatric populations of the two species seem to exist in this region (Gallego, 2001, Kerdelhué, 2002, Kohlmayr, 2002). The two species are morphologically difficult to distinguish, even when there are obvious differences in their life cycle and genetic structure (Gallego, 2001, Kerdelhué, 2002, Kohlmayr, 2002).

Host preference has often been described as a way to isolate specialized populations that eventually leads to sympatric speciation (Tauber and Tauber, 1989; Bush and Smith, 1997). The two sibling species are expected to infest, or at least prefer, different *Pinus* species (Pfeffer, 1995, Kerdelhué, 2002). However, our field observations indicate that the two species seem to live in sympatry in Portugal, both colonizing maritime pine *Pinus pinaster* (Vasconcelos *et al.*, 2005).

The goal of the present work is to analyse the host preferences of *T. piniperda* and *T. destruens* as possible evidence for the role of the host plant in speciation. Improved knowledge of the geographical distribution of the two sibling species is a further objective.

II. Material and Methods

A. Beetle sampling

Four *Tomicus* populations were sampled (Fig. 1, Table 1): 1) In September/October 2002, mature beetles were sampled from *Pinus pinaster* shoots in Vila Pouca de Aguiar. In January parent beetles were sampled from different gallery systems in the same place and host trees.

2) In October 2002 mature beetles were sampled from *Pinus pinea* shoots in Alcácer do Sal.

3) In October 2002, parent beetles were sampled from *Pinus pinaster* trap logs in Ota.

4) In January 2003, parent beetles attacking living *Pinus pinaster* trees were sampled in Ponte de Lima.

About 250 insects were sampled from each population. Twenty of them were immediately killed and stored in absolute ethanol to genetically determine which species of *Tomicus* they belonged to. The other ones were kept alive to test host preference.



1-Ponte de Lima *Pinus pinaster* trees

2-Vila Pouca de Aguiar *Pinus pinaster* shoots *Pinus pinaster* logs

3-Ota Pinus pinaster logs

4-Alcácer do Sal *Pinus pinea* shoots

Fig. 1. Location of the experimental stands

		Mean	Mean	Mean
Site	Geographical	annual	annual	temperature
	coordinates	temperature	rainfall	range
		(°C)	(mm)	(°C)
Ponte de	41°46' N	14.2	1720.1	9.5 (Jan)
Lima	8°36' W	14.5	1/20.1	20.0 (Jul)
Vila Pouca de Aguiar	41°30' N	13.4	1503.7	6.4 (Jan)
	7°39' W			21.4 (Jul)
Ota	39°07' N	16.1	587.3	10.2 (Jan)
	8°59' W			22.4 (Aug)
Alcácer do	38°23' N	16.2	574 5	10.3 (Jan)
Sal	8°31' W	10.5	574.5	23.0 (Aug)

TABLE I Sampling site characteristics

B .*Host preference tests*

Beetles collected from shoots were held at 4°C inside the shoots in a growth chamber until they were used in choice tests. Beetles collected from logs were carefully removed from the bark immediately before use. Only active *Tomicus* adult females with full legs and antennae were used for host preference tests. Females were determined by their inability to stridulate.

Maritime pine (MP), *Pinus pinaster*, Aleppo pine (AP), *Pinus halepensis*, and stone pine (SP), *Pinus pinea*, logs (diameter: 6-12 cm, length: 30 cm) were cut from three sites in Portugal in September 2002 (tests conducted in October 2002) and in December 2002/January 2003 (tests conducted in January 2003). The ends of the logs were coated with paraffin to avoid desiccation. For each test, two logs of different species with similar diameter and bark characteristics were paired in a bag and one beetle was placed between them. Three or four days after, host choice (evaluated by initiation of galleries), the number of dead beetles and the number of missing beetles were recorded. All possible pair combinations were tested.

C. Data analysis

Data was analysed by hierarchical log-linear models (SPSS 10.0 Software). The dependent variable was the number of times each species was selected. The explanatory variables were two factors, the pine shoot beetle population and the host species, and their interaction. A goodness-of-fit statistic, the Likelihood ratio chi-square (LRChisq), was used to assess model adequacy. A model was considered adequate if P > 0.05. Considering the principle of parsimony, the model with the fewest variables yet adequate goodness-of-fit was selected. The best model was selected by backward elimination of variables ($\alpha = 0.05$) starting from the saturated model (P = 1.00).

III. Results and Discussion

All beetles from Ota, Alcácer do Sal and Ponte de Lima were identified as *Tomicus destruens*. On the other hand, beetles from Vila Pouca de Aguiar (Northeast of Portugal) were identified as *Tomicus piniperda*, which is probably related to the fact that this site has a more continental climate with the lowest winter temperature (Table 1).

This is the first unequivocal demonstration of the large presence of *T. destruens* in Portugal, suggesting that this species is dominant in this region while *T. piniperda* is present in lower numbers. Until now only two species of the genus *Tomicus*, *T. piniperda* and *T. minor*, were reported in Portugal (Ferreira, 1998, Ferreira and Cabral, 1999).

Concerning host preference, on both choice tests for MP \times SP and MP \times AP, there was no significant effect detected for the beetle population factor was found. The best model only included the host effect as a significant factor, LRChisq = 1.61, df = 8, P = 0.991 and LRChisq = 13.09, df = 8, P = 0.109, respectively. In both cases MP was the preferred host, chosen by 70% of the beetles when compared with SP and by 60% when compared with AP (Fig. 2 and 3).



Fig. 2. *Tomicus* preference between maritime pine (MP) and stone pine (SP). Different bars show the number of beetles released and the number of beetles that initiated galleries in each host species.



Fig. 3. *Tomicus* preference between maritime pine (MP) and Aleppo pine (AP). Different bars show the number of released beetles and the number of beetles that initiated galleries in each host species.

These results suggest that for both species of *Tomicus* and all the populations we tested, MP is a very attractive host, preferred to the two other Mediterranean pine species. Maritime pine is the dominant pine species in Portugal, covering about 90% of the pine stands in the country. A regional adaptation of *Tomicus* populations to the local dominant pine species may have occurred. Siegert and McCullough (2001) observed similarly that *T. piniperda* sampled in North America prefers Scots pine, *Pinus sylvestris*, one of the main pine hosts in this region. On the other hand, our results do not agree with the hypothesis that *T. piniperda* and *T. destruens* infest different pine species (Kohlmayr, 2002). Furthermore, they indicate that host preference is probably independent of maternal effects as we observed that beetles sampled from a SP stand still preferred MP.

For the AP × SP tests, the beetle population, the host tree species and their interaction were all significant effects. Further analysis showed that the two *Tomicus* populations from South Portugal (Ota and Alcácer do Sal) preferred SP (LRChisq = 0.49, df = 2, P = 0.783) while the two populations from North Portugal (Vila Pouca de Aguiar and Ponte de Lima) preferred AP (LRChisq = 3.37, df = 2, P = 0.185) (Fig. 4).



Fig. 4. *Tomicus* preference between Aleppo pine (AP) and stone pine (SP). Different bars show the number of beetles released and the number of beetles that initiated galleries in each host species.

Therefore intraspecific differences may exist between *Tomicus* populations regarding their host preference. Our results are not very conclusive about such differences between *T. piniperda* and *T. destruens*, and further experiments are necessary on this subject.

We found one population of T. destruens (Ponte de Lima) breeding in January, although this species is supposed to breed during autumn (Gallego, 2001). One explanation could be the existence of T. destruens sister broods that extended through the winter.

This preliminary work has provided interesting results, but further work will be needed to clarify life cycle differences between *T. piniperda* and *T. destruens*.

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