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POPULATION DYNAMICS AND SEASONAL ABUNDANCE OF *THAUMASTOCORIS PEREGRINUS* IN *EUCALYPTUS* PLANTATIONS IN SARDINIA (ITALY) ⁽¹⁾

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Deiana V., Pinna C., Buffa F., Mannu R., Satta A., Floris I. – Population dynamics and seasonal abundance of *Thaumastocoris peregrinus* in *Eucalyptus* plantations in Sardinia (Italy)

Thaumastocoris peregrinus is a eucalyptus pest native to Australia. It is currently the only species belonging to the Thaumastocoridae family in Europe. In Italy, it was reported for the first time in Latium in 2011, whereas in Sardinia it was detected in January 2015 on a *Eucalyptus camaldulensis* plantation located in the south of the island. Our research team carried out a monitoring program for two-years in order to verify its geographical distribution, the main periods of infestation, the flight peaks of the adults and to also detect the presence of unhatched eggs on leaves. During the first year, the occurrence of adults of *T. peregrinus* was verified by yellow sticky traps placed in twelve areas distributed throughout the island where *Eucalyptus* plantations were located. In the second year, the population dynamics and the occurrence of eggs on leaves were evaluated only in three representative study areas, already included in the previous monitoring program, located in the north, centre and south of the island, respectively. Field surveys showed the presence of *T. peregrinus* in all the monitored locations. The highest adult population was observed from late summer to initial autumn, with a significant increase in adults caught by yellow sticky traps from August, which reached its peak in September and gradually decreased in the following months. The presence of eggs of *T. peregrinus* on leaves from June to December also indicates that the insect is currently well established in the region.

KEY WORDS: bronze bug, *Eucalyptus* pest, nonnative species, monitoring.

INTRODUCTION

The bronze bug *Thaumastocoris peregrinus* Carpintero et Dellapé (Hemiptera Thaumastocoridae) is a sap-feeding insect native to Australia, which infests plant species belonging to Myrtaceae (NOACK *et al.*, 2011). Its common name is due to the symptoms associated with infestation: adults and nymphs feeding on leaves lead to a decrease in the photosynthetic area and leaf discoloration, followed by leaf silvering and tanning (DIAS *et al.*, 2014).

Bronze bug infestations cause tree defoliation, branch dieback, and reduction in tree growth, as well as tree death when attacks are particularly severe and prolonged (JACOBS and NESER, 2005; NADEL *et al.*, 2015). *T. peregrinus* thus represents a potential key pest of different plant species, especially in areas where the insect has been newly introduced.

The bronze bug has been recently found in several areas outside its native range, such as South Africa (JACOBS and NESER, 2005), Argentina (NOACK and COVIELLA, 2006), Uruguay (MARTÍNEZ and BIANCHI, 2010), Brazil (WILCKEN *et al.*, 2010), Italy (LAUDONIA and SASSO, 2012), Portugal (GARCIA *et al.*, 2013), and Mexico (JIMENEZ-QUIROZ *et al.*, 2016). Mediterranean and subtropical areas are considered

to be the most suitable for *T. peregrinus* worldwide, as already observed by the analysis of its potential global distribution (MONTEMAYOR *et al.*, 2015; SAAVEDRA *et al.*, 2015). Among all potential hosts, *T. peregrinus* has been generally associated with several *Eucalyptus* species (LAUDONIA and SASSO, 2012; SOLIMAN *et al.*, 2012; GARCIA *et al.*, 2013), and different development rates and levels of infestation have been observed among the plant species on which it develops (JACOBS and NESER, 2005; NOACK and COVIELLA, 2006; IDE *et al.*, 2011; SOLIMAN *et al.*, 2012; BARBOSA *et al.*, 2014; SANTADINO *et al.*, 2017).

SOLIMAN *et al.* (2012) showed that the longevity of bronze bug adults varies from 4 to 78 days, and males generally live longer than females. On average, *E. urophylla* and *E. grandis* were found to be the most suitable species for the development and reproduction of *T. peregrinus* under laboratory conditions (SOLIMAN *et al.*, 2012). However, *T. peregrinus* develops well on other *Eucalyptus* species, such as *E. camaldulensis* (LAUDONIA and SASSO, 2012; SOLIMAN *et al.*, 2012; GARCIA *et al.*, 2013), thus leading to severe infestations and serious damages (JACOBS and NESER, 2005; GARCIA *et al.*, 2013).

E. camaldulensis is the most important *Eucalyptus* species cultivated in Italy, and is used as a windbreak and as ornamental plants in parks and gardens, as well as being cultivated for biomass fuel. In Italy, *Eucalyptus* plantations are located above all in central-southern regions, including Sardinia and Sicily (DEIDDA *et al.*, 2016).

In Sardinia, *E. camaldulensis* was introduced at the beginning of the last century and primarily in land

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reclamation areas. Today, *Eucalyptus* plantations are predominantly at less than 400 m above sea level (DEIDDA *et al.*, 2016), covering approximately 23,000 hectares and representing one of the main regions for *Eucalyptus* plantations in Italy (GASPARINI and TABACCHI, 2011). *Eucalyptus* trees are commonly found in several agricultural and forest landscapes in Sardinia and their flowers are valuable sources of nectar and pollen (FLORIS *et al.*, 2007). The presence of *T. peregrinus* in such areas has a negative impact on the phytosanitary status of *Eucalyptus*, which has already been affected by several phytophagous species such as psyllids, particularly the red gum lerp psyllid *Glycaspis brimblecombei* Moore (Hemiptera Aphalaridae) (DEIDDA *et al.*, 2016; MANNU *et al.*, 2018).

A monitoring network in Sardinia covering different *Eucalyptus* distribution areas was carried out to: 1) obtain information on the distribution of *T. peregrinus* in Sardinia; 2) evaluate the seasonal abundance and main periods of infestation of bronze bug under Mediterranean conditions. We had already conducted similar study for *G. brimblecombei* (FLORIS *et al.*, 2018; MANNU *et al.*, 2018).

MATERIALS AND METHODS

FIELD SURVEYS AND SAMPLING PROCEDURE

Field surveys were carried out in 2015 and 2016, and stations were selected to cover the northern, central and southern parts of Sardinia. In 2015, four stations for each macroarea for a total of 12 were monitored, and three stations (one for each macroarea) were surveyed in 2016 (Fig. 1): “Santa Maria La Palma” (Northern), “Ottana” (Central) and “Uta” (Southern). In both years we monitored the “Uta” location as it was the nearest to the first observation site of *T. peregrinus* in Sardinia (DI LASCIO and NANNINI, 2016).

Adults of *T. peregrinus* were monitored in 2015 and 2016 using yellow sticky traps, which are considered to be one of the most effective methods for capturing adults (NADEL *et al.*, 2015). Four yellow sticky traps (20 × 20 cm) were placed in four different *Eucalyptus* trees randomly selected at each location. Traps were positioned on branches located in the middle of the canopy, which is the best position to capture *T. peregrinus* adults (MARTÍNEZ *et al.*, 2010; NADEL and NOACK, 2012; NADEL *et al.*, 2015).

Fig. 1 – Distribution of locations constituting the monitoring network of *T. peregrinus* adults and eggs in Sardinia (Italy). Numbers from 1 to 12 indicating each location are sorted according to a North-South gradient (1=“Olbia”; 2=“Santa Maria La Palma”; 3=“Ozieri”; 4=“Alghero”; 5=“Ottana”; 6=“Siniscola”; 7=“Arbatax”; 8=“Arborea”; 9=“Serramanna”; 10=“San Vito”; 11=“Siliqua”; 12=“Uta”). Different colors in the map indicate different geographical areas (Orange=North Sardinia; Green=Central Sardinia; Grey=South Sardinia).

The traps were collected and replaced biweekly in the summer and monthly in other seasons. The traps collected in the field were then taken to the laboratory where the *T. peregrinus* adults were counted

Observations of eggs were conducted only in 2016. At each location and sampling date, four branchlets of approximately 40 cm in length were collected from the canopy of the four trees. Again, samplings were carried out from the middle of the canopy, as it is also recommended for assessing the density of bronze bug eggs (MARTÍNEZ *et al.*, 2010; NADEL *et al.*, 2015; JIMENEZ-QUIROZ *et al.*, 2016). Each branchlet was collected separately in a plastic bag and transferred to the laboratory, where the numbers of unhatched eggs were counted under a stereoscopic microscope on both sides of four mature leaves per branch.

STATISTICAL ANALYSIS

All statistical analysis were performed using R software (R CORE TEAM, 2016). Firstly, abundance data were $\log(x+1)$ -transformed to satisfy the normality assumption. In order to explore the effects of location on the abundance of *T. peregrinus* adults and eggs, a Linear Mixed Model (LMM) was considered for each year using the lmer function of the “lme4” package in R (BATES *et al.*, 2015). Location was considered as the fixed factor, whereas sampling dates and traps were the random factors. An LMM was fitted to the 2015 monitoring data to evaluate the effects of the different geographical areas on the abundance of *T. peregrinus* adults, considering the geographical areas as the fixed factor and sampling dates and traps as random factors. Analysis of variance (ANOVA) considering Type II Wald F tests with Kenward-Roger correction of degrees of freedom was used to test the significance of factors, followed by Tukey’s post-hoc test at a significance level of $p < 0.05$ for mean separation.

Finally, a linear regression model approach was used to evaluate the relationship between the average number of

eggs and adults captured either at the same or different sampling times. The average monthly numbers of eggs observed in 2016 in all locations were thus compared to those of adults captured either in the same (t), or previous ($t-1$), or following ($t+1$) month, by linear regression models. ANOVA was performed to test the significance of the regression models and Pearson’s correlation coefficient was calculated to evaluate the relationship between the variables.

RESULTS

Adults of *T. peregrinus* were captured in all monitored locations in 2015, and statistical differences among locations were found ($F_{11,40.257}=20.30$; $p < 0.001$). “Serramanna”, “Uta”, “Olbia” and “Arbatax” were the most infested locations, which showed an average monthly number of adults greater than 10 individuals per trap (Table 1). However, five locations showed average values lower than one adult per month compared to all the monitored areas. Statistical differences in adult abundance were also found between geographical areas ($F_{2,73.158}=7.37$; $p < 0.001$). The highest and the lowest average monthly number of adults captured by traps was observed in south (16.3 ± 3.1) and north (4.2 ± 1.7) Sardinia, respectively. The seasonal abundance of *T. peregrinus* in 2015 was similar in all the geographical areas (Fig. II). The population dynamics of adults was comparable in the three geographical areas, showing an increase in the average number of adults in August and a peak in population abundance from September to October in southern and central Sardinia. Although population abundance increased simultaneously in all areas, in northern Sardinia, the number of adults decreased immediately after September.

Significant differences between the three areas were also found in 2016 both for adults ($F_{2,9}=14.96$; $p < 0.001$) and eggs ($F_{2,9}=14.60$; $p < 0.001$). The “Ottana” (i.e. central Sardinia) location showed the highest average monthly

Table 1 – Average monthly number of adults per traps and eggs per branch of *T. peregrinus* captured at 12 and 3 locations distributed throughout Sardinia (Italy) during 2015 and 2016, respectively. Labels identify each location according to North-South gradient. Values are reported as mean $\pm$ standard error. Different letters in a column indicate significant differences in abundance among locations within same year (Tukey’s post hoc test, $p < 0.05$).

Label	Geographical area	Location	2015	2016	
			Adults	Adults	Eggs
1	North Sardinia	Olbia	12.45 $\pm$ 3.90 b	-	-
2	North Sardinia	S. M. La Palma	0.02 $\pm$ 0.02 a	8.36 $\pm$ 2.58 ab	0.58 $\pm$ 0.34 a
3	North Sardinia	Ozieri	0.09 $\pm$ 0.09 a	-	-
4	North Sardinia	Alghero	0.02 $\pm$ 0.02 a	-	-
5	Central Sardinia	Ottana	0.03 $\pm$ 0.02 a	20.54 $\pm$ 5.03 b	7.05 $\pm$ 1.81 b
6	Central Sardinia	Siniscola	6.38 $\pm$ 1.97 b	-	-
7	Central Sardinia	Arbatax	11.11 $\pm$ 4.00 b	-	-
8	Central Sardinia	Arborea	0.03 $\pm$ 0.02 a	-	-
9	South Sardinia	Serramanna	16.67 $\pm$ 5.51 b	-	-
10	South Sardinia	San Vito	8.72 $\pm$ 3.08 b	-	-
11	South Sardinia	Siliqua	7.16 $\pm$ 2.61 b	-	-
12	South Sardinia	Uta	16.48 $\pm$ 4.74 b	4.82 $\pm$ 1.28 a	0.21 $\pm$ 0.07 a

Fig. II – Average number of adults of *T. peregrinus* captured monthly during May–December 2015 in South, Central and North Sardinia (Italy). Error bars in graph indicate standard error of the average number of adults per month.

values of both adults and eggs compared to the other areas (Table 1). The seasonal abundance of eggs over time was comparable to that of adults in each location (Fig. III). Despite this, adults and egg abundance in “Ottana” reached their peak in August, whereas in other locations an increase in the population of both adults and eggs was observed only after the middle of September. In general, the seasonal abundance of eggs was comparable to that of adults in the same location, with an earlier abundance peak of eggs than of adults.

Finally, the average monthly number of eggs was significantly related to the average number of adults captured in the same month ($F_{1,29}=29.73$; $p<0.001$; Pearson’s $R=0.72$) and in the following month ($F_{1,26}=59.49$; $p<0.001$; Pearson’s $R=0.84$). On the other hand, no significant relationship was found between eggs and the average number of adults captured in the previous month ($F_{1,26}=0.62$; $p=0.43$; Pearson’s $R=0.16$) (Fig. IV).

DISCUSSION

T. peregrinus is a serious pest of *Eucalyptus* species worldwide (LAUDONIA and SASSO, 2012; SOLIMAN *et al.*, 2012). The presence of the pest in all monitored locations throughout Sardinia ten months after its first recording confirms that the Mediterranean basin is one of the most suitable areas for its settlement (LAUDONIA and SASSO, 2012; MONTEMAYOR *et al.*, 2015; SAAVEDRA *et al.*, 2015). The rapid diffusion of *T. peregrinus* in Sardinia after its introduction may be due to both its high passive dispersal ability through wind and human transport (WYLIE and SPEIGHT 2012; SASSO *et al.*, 2014), and its capacity to remain active in the field during all seasons (SASSO *et al.*, 2014; NADEL *et al.*, 2015). In Sardinia, the highest population abundance of bronze bug was also observed during the seasonal period characterized by the highest presence of tourists (from August to September), which probably represents one of the most important phoretic means for its spread. However, the insect may also spread during other seasons, as no diapause period has been

observed for *T. peregrinus* (NADEL *et al.*, 2015). The spread of *T. peregrinus* adults in the field on a local scale during all seasons, is also probably positively affected by the emission of different volatile compounds from uninfested trees, which are presumed to act as kairomones on mated females (CAMILA *et al.*, 2013).

The adult population dynamics observed during the first monitoring year in Sardinia was comparable to that reported for other infested regions in Italy (SASSO *et al.*, 2014). In 2015, the adult dynamics followed the same pattern in all three areas in Sardinia, despite the different environmental conditions of the monitored locations (DEIDDA *et al.*, 2016; MANNU *et al.*, 2018). However, a few differences in population dynamics, as well as seasonal abundances, were found from area to area in the second year. Although in southern Sardinia seasonal abundances decreased from the first to the second year, the adult population dynamics did not show a different pattern compared to 2015. In contrast to this, an increase in seasonal abundance was observed both in central and northern Sardinia, likely indicating the spatial expansion of *T. peregrinus* population throughout the Island, in line with the typical pattern of an invasive alien (non native) species (ANDOW *et al.*, 1990). In fact, biological invasions are always characterized by three time-steps (1) introduction of the invasive species, (2) its establishment, and (3) spatial expansion into new areas (ELTON, 1958).

In addition, the adult dynamics recorded in 2016 in central Sardinia was different to those of northern and southern Sardinia observed in the same and previous years. Population dynamics and seasonal abundance are generally affected by different factors, including management and environmental conditions, which both directly and indirectly influence other *Eucalyptus* pest species (LAUDONIA *et al.*, 2014; MANNU *et al.*, 2018). Of these, water availability, especially during the summer, is considered one of the most influencing factors as it allows a good foliage development which might influence sucking insect feeding (LAUDONIA *et al.*, 2014).

Finally, the high correlations obtained between the average number of eggs and the average number of adults

Fig. III – Average number of (1) unhatched eggs per branch and (2) adults per trap of *T. peregrinus* observed at each sampling date during June-December 2016 in Ottana (Central Sardinia), Santa Maria La Palma (Northern Sardinia), and Uta (South Sardinia), respectively.

could be particularly useful for improving monitoring activities. They could enable adult population abundance of the bronze bug to be predicted in the short-term, particularly from the perspective of biological control, e.g. using the specific egg parasitoid *Cleruchoides noackae* Lin and Huber (Hymenoptera: Mymaridae) (BARBOSA *et al.*, 2017).

CONCLUSIONS

In Sardinia *T. peregrinus* is now well established throughout the island just two years after its introduction. The presence of bronze bug complicates the health of *Eucalyptus* plantations, which are already being affected by the presence of various phytophagous and several phytopathogens (DEIDDA *et al.*, 2016). This highlights the need to take phytosanitary measures to contain and limit the entry of other new species which could lead to irreversible ecological changes and economic losses. In accordance with Italian legislation, we are now evaluating the possibility of introducing the specific egg parasitoid *C. noackae*, due to its promising potential for the biological control of *T. peregrinus* (BARBOSA *et al.*, 2017).

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