

**FIRST RECORD OF *QUADRASTICHUS MENDELI*, A PARASITOID OF
LEPTOCYBE INVASA IN SOUTH AFRICA**

Samantha J. Bush^{1,3}, Gudrun Dittrich-Schröder^{1,3}, Stefan Naser³, Caitlin Gevers^{1,3}, Kwabena Owusu Baffoe^{1,3}, Bernard Slippers^{2,3} and Brett P. Hurley^{1,3}

¹ *Department of Zoology and Entomology, University of Pretoria, Pretoria, 0002, South Africa*

² *Department of Genetics, University of Pretoria, Pretoria, 0002, South Africa*

³ *Forestry and Agricultural Biotechnology Institute, University of Pretoria, Pretoria, 0002, South Africa*

Corresponding author: Samantha.Bush@fabi.up.ac.za

Leptocybe invasa, the blue gum chalcid, is a well-known pest of *Eucalyptus*. Since it was first noticed in the Mediterranean and Middle East in 2000, it has spread to other *Eucalyptus* growing countries around the world. Efforts to control it have included the release of a number of larval and pupal parasitoids. This Report Note serves as the first record of the presence of *Quadrastichus mendeli* in South Africa, a parasitoid originally released as a biological control agent of *L. invasa* in Israel. The interactions and potential impacts on the other insects in the *L. invasa* galls will need to be investigated.

Keywords: biological control, *Eucalyptus*, gall wasp, Hymenoptera, invasive pest

INTRODUCTION

The increase in global movement of goods and people in recent years has seen an increase in insect movement and establishment (McCullough et al. 2006; Aukema et al. 2010; Garnas et al. 2012). This is particularly so with insect pests of *Eucalyptus* species, where the rate of introduction of new non-native pests across the globe has increased five-fold since 1986 (Hurley et al. 2016). The blue gum chalcid, *Leptocybe invasa* Fisher & La Salle (Hymenoptera: Eulophidae), is one such example. This gall-forming insect, thought to originate from Australia, was first noticed as a pest on *Eucalyptus* in the Mediterranean and Middle East in 2000 (Mendel et al. 2004). Since then, this pest has spread to many countries in Africa, Asia, Europe and North and South America (e.g. Mendel et al. 2004; Dhahri and Ben Jamaa 2010; Mifsud 2012; Diaz et al. 2014; Vanegas-Rico et al, 2015). *Leptocybe invasa* was first reported in South Africa in 2007 (Kelly et al. 2012) and is currently considered one of the most serious insect pests of *Eucalyptus* in the country.

Classical biological control, the introduction of natural enemies from the native range of a pest to control a pest organism in its invaded range (Eilenberg et al. 2001), has been one of the main approaches to manage populations of *L. invasa* in South Africa and abroad. This effort was led by researchers in Israel, where *Selitrichodes kryceri* Kim & La Salle (Hymenoptera: Eulophidae), *Quadrastichus mendeli* Kim & La Salle (Hymenoptera: Eulophidae), *Megastigmus lawsoni* Doğanlar (Hymenoptera: Torymidae) and *M. zvimendeli* Doğanlar (Kim et al. 2008; Doğanlar and Hassan 2010) were released. *Quadrastichus mendeli* has subsequently been sent to India from Israel (Jacob et al. 2015). In addition to this deliberate movement of *Q. mendeli* it has also been reported in Italy as an unintentional introduction (Nugnes et al. 2016).

A separate survey in Australia, by South African researchers, resulted in the collection and identification of another parasitoid of *L. invasa*, namely *Selitrichodes neseri* Kelly & La Salle (Hymenoptera: Eulophidae) (Kelly et al. 2012). *Selitrichodes neseri* was released in South Africa in 2012 (Dittrich-Schröder et al. 2014) and subsequently in Mauritius, Brazil, Zimbabwe and Chile (authors, unpublished).

Other than *S. neseri*, no biological control agents of *L. invasa* have been intentionally released in South Africa. An unidentified wasp, that was neither *S. neseri* nor the *Megastigmus* species that inhabit *L. invasa* galls, namely *M. zebrinus* Grissell (Hymenoptera: Torymidae) and *M. pretorianensis* Doğanlar (Hymenoptera: Torymidae) emerged from galled material collected in South Africa. This Research Note reports the identification of this wasp based on morphological characteristics and DNA sequence data. Preliminary data is provided on the current distribution of this species in the country.

METHODS AND RESULTS

Galled *Eucalyptus* material was collected from the *Eucalyptus* plantation of the National Zoological Gardens of South Africa at Rietondale, Pretoria, South Africa (25°43'58.50"S 28°14'20.26"E) in March 2016. This material was kept in 9 litre, 255 x 150 x 260mm, rectangular plastic containers at the Forestry and Agricultural Biotechnology Institute (FABI) Biocontrol Facility at the Experimental Farm of the University of Pretoria. Emerging wasps were collected and separated morphologically as *L. invasa*, *S. neseri*, *M. zebrinus* and *M. pretorianensis*. Amongst this collection were two individuals that did not belong to any of the known inhabitants of *L. invasa* galls in South Africa.

Morphological characteristics described by Kim et al. (2008) were used to tentatively identify the hymenopteran. Specific characteristics included a small body size (1.15-1.35mm), a mainly yellow body with dark brown markings, antennae with three funicular segments longer than they are wide and a large anellus, as well as a gaster longer than the head plus mesosoma. These unknown individuals were tentatively identified as females of *Quadrastichus mendeli* (Figure 1), and later confirmed to be this species by O. C. Neser (formerly of Biosystematics, ARC-PPRI, Pretoria) by comparison with specimens received from Israel.



Figure 1 (a) Female *Leptocybe invasa*, (b) galls on a *Eucalyptus* sp. branch, (c) dissected gall showing multiple developing *L. invasa*, and (d) *Quadrastichus mendeli* adult.

The morphological identification was confirmed by sequencing a portion of the cytochrome *b* gene of the mitochondrial DNA (mtDNA) of one of the collected female specimens (GenBank, reference number KX757846) and comparing that to a confirmed *Q. mendeli* specimen from Israel (GenBank reference number KX757847). The primers CP1 (5'-GAT GAT GAA ATT TTG GAT C-3') (Harry et al. 1998) and CB2 (5'-ATT ACA CCT CCT AAT TA TTA GGA AT-3') (Jermin and Crozier 1994) were used to amplify and sequence the cytochrome *b* locus. The sequence results were identical to each other and thus confirmed the specimen as *Q. mendeli*.

Material galled by *L. invasa* has been collected from the *Eucalyptus* plantation of the National Zoological Gardens approximately twice a month during spring and summer since 2013 and the insects that have emerged screened. Until March 2016, *Q. mendeli* had not been collected from the samples. *Leptocybe invasa* galled material collected in 2016 as part of a national *L. invasa* monitoring initiative in South Africa was screened to determine if *Q. mendeli* was present in other areas of South Africa. As this was a pilot study for this monitoring initiative, only a limited amount of material was collected (87 trees across 19 sites in Mpumalanga, Limpopo and KwaZulu-Natal). *Quadrastichus mendeli* was not detected from material collected at any of these sites (authors, unpublished data). In addition to the monitoring initiative, *Q. mendeli* was also not found in *L. invasa*-infested material collected during a two year study (2012/2013) that monitored the establishment success of *S. nesei* in KwaZulu-Natal, Mpumalanga and Limpopo. However, recently it was discovered close to Buffelspoort, North West Province (25° 49.866'S, 27° 24.676'E).

DISCUSSION

This study provides the first report of *Q. mendeli* in South Africa and in sub-Saharan Africa. *Quadrastichus mendeli* is a parasitoid of young and mature *L. invasa* larvae and it is currently unknown whether the larvae are killed immediately or shortly after parasitization (Kim et al. 2008). *Quadrastichus mendeli* is a uniparental and solitary parasitoid with an approximate developmental time of 30 days at 28.1°C (Kim et al. 2008). Laboratory studies in Israel have shown a parasitism rate of between 7.9 and 84%, depending on the life stage attacked (Kim et al. 2008). Results from field monitoring in Italy have shown a parasitism rate of between 30.2 and 50.5% (Nugnes et al 2016). The short life cycle, compared to that of *L. invasa*, is thought to provide an advantage as a biological control agent (Nugnes et al. 2016).

Leptocybe invasa material has been extensively collected since its first appearance in 2007. The absence of *Q. mendeli* in South Africa from this previously collected material indicates a limited distribution and suggests that the parasitoid has only recently been introduced into the country. Future monitoring is needed to determine the prevalence and distribution of this parasitoid.

Kim et al. (2008) showed that *Q. mendeli* is an ectoparasitoid of *L. invasa* and raised the possibility that *Q. mendeli* could also develop on other *Leptocybe* species in Australia. Indeed, it is not known whether or not *Q. mendeli* parasitizes other occupants of *L. invasa* galls, namely the *Megastigmus* spp. and the biological control agent *S. neseri*. Studies are therefore needed to determine the impact of this new parasitoid on populations of *L. invasa*, as well as the possible interactions associated with these galls, especially the biological control agent *S. neseri*.

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