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Host specificity of different populations of the leaf beetle *Diorhabda elongata* (Coleoptera: Chrysomelidae), a biological control agent of saltcedar (*Tamarix* spp.) ☆

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Abstract

The leaf beetle, *Diorhabda elongata* (Brullé) *sensu lato*, was released in 2001 for the classical biological control of exotic saltcedars, a complex of invasive *Tamarix* species and hybrids. It did not establish at sites south of 37°N latitude where summer daylengths are below the critical photoperiod of the northern-adapted populations of the beetle that were released. Therefore, we assessed the host specificity of four *D. elongata* populations collected from more southern latitudes in the Old World (Tunisia, Crete, Uzbekistan, and Turpan, China). All populations were similar to each other and the previously released populations of *D. elongata* in their host specificity. Larval/pupal survival for all populations was 34–100% on *Tamarix* test plants, 0–76% on native *Frankenia* plants (both in the order Tamaricales), and 0% on the remaining 28 species of plants on which all the larvae died as 1st instars. *D. elongata* laid high numbers of eggs on saltcedar, generally fewer eggs on athel (a moderately valued evergreen species of *Tamarix*) except for Uzbekistan beetles, and few to no eggs on three species of *Frankenia*. Few to no adults were found on *Frankenia* plants which also were poor maintenance hosts. The release of any of the four *D. elongata* populations in the southern US and northern Mexico should pose no risk to plants outside the order Tamaricales and a low risk to native, non-target *Frankenia* plants. Athel may be less damaged than saltcedar.
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1. Introduction

Saltcedars (*Tamarix* spp., Tamaricales: Tamaricaceae) are deciduous shrubs or small trees of riparian areas in deserts and steppes of Eurasia and Africa (Baum, 1978). Ten species of *Tamarix*, including nine different saltcedars and the single evergreen species *T. aphylla* (L.) Karsten (athel), were introduced into the United States and Mexico beginning in the early 1800s, primarily as ornamentals, for windbreaks and shade, and to stabilize stream banks (Baum, 1967; Crins, 1989; DiTomaso, 1998). Following the late 1920s, some of the saltcedar species became highly invasive along western riparian areas and lakeshores, with an early estimate of 600,000 ha of bottomlands infested (Robinson, 1965). The primary species involved in this invasion are *Tamarix ramosissima* Ledebour and *T. chinensis* Loureiro, as well as a common and widespread hybrid that has formed between these two species (Gaskin and Schaal, 2002). Additional invasive taxa include *T. parviflora* de Candolle, *T. canariensis* Willdenow, *T. gallica* L. (the latter two species being difficult to distinguish), and hybrids involving combinations of *T. ramosissima* and *T. chinensis* with *T. parviflora* and *T. canariensis*/*T. gallica* (Gaskin and Schaal, 2002, 2003). Saltcedar infestations currently range

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from North Dakota to Washington and south to northern Mexico. Both anthropogenic changes in western riparian ecosystems, which created ideal conditions for saltcedar invasion of disturbed areas, as well as the ability of saltcedar to invade and modify undisturbed environments, has created what is considered to be an ecological disaster for riparian areas of the West (DeLoach et al., 2000).

The *Diorhabda* leaf beetle from Fukang, China and Chilik, Kazakhstan, designated as *Diorhabda elongata* (Brullé) *deserticola* Chen by DeLoach et al. (2003) and by Lewis et al. (2003a,b), was released into the open field in 2001 in the United States for the classical biological control of saltcedar. Both adults and larvae defoliate saltcedar; 3rd instars are the most damaging stage (Lewis et al., 2003b). These beetle populations are established and increasing in abundance at most sites north of 37°N latitude (DeLoach et al., 2004). However, no establishment occurred at more southern sites where summer day lengths are less than 14 h 30 min, which is the critical photoperiod for the Fukang population (D.W. Bean, personal communication). In the southern areas, adult beetles enter reproductive diapause in early summer and presumably deplete their fat body reserves and starve before the following spring (Lewis et al., 2003b). This left large areas of some of the most serious saltcedar infestations, from Texas and Oklahoma west to southern California, without a biological control agent.

Other, more southern, populations of *D. elongata sensu lato* have shorter critical photoperiods for diapause induction (DeLoach et al., 2004; D.W. Bean, personal communication) and therefore are more likely to establish at more southern latitudes in North America. However, variation in this and other biological traits among populations of *D. elongata*, especially those distant from the original source of beetles released, may be accompanied by variation in their host range as well. A primary concern in the saltcedar biological control program has been assessing the risk posed to native, non-target plants in the genus *Frankenia* L., small shrubs of desert and salt marsh habitats (Lewis et al., 2003a). In North America, six species occur in the southwestern United States and northern Mexico (Whalen, 1980, 1987). The genus *Frankenia* is placed in the Frankeniaceae and together with the Old World family Tamaricaceae comprises the order Tamaricales (Spichiger and Savolainen, 1997). *Frankenia* spp. are the only native members of the Tamaricales found in North America (or even the Western Hemisphere); *Tamarix* is the only exotic genus of the order present (Whalen, 1980, 1987). A secondary concern involves the introduced athel, which is native to parts of southern Asia and northern and eastern Africa (Baum, 1978). It is a cold-intolerant, tree-sized, evergreen species of *Tamarix* that is grown as a drought-tolerant shade tree and windbreak, especially in northern Mexico.

We report here our evaluation of the host specificity of four populations of *D. elongata* collected across a wide geographical area of Eurasia and North Africa and below 43°N latitude, three of which had never been tested previously. This information, in combination with other biological studies of candidate populations, will provide the basis for determining the most promising population of *D. elongata* to release in the southern areas of the saltcedar infestation in North America.

2. Materials and methods

2.1. Insect colonies

The *Diorhabda* beetles collected on *Tamarix* in Asia and the Mediterranean area were all identified as *D. elongata* by A.S. Konstantinov (USDA-Agricultural Research Service Systematic Entomology Laboratory, Beltsville, MD) and/or I.K. Lopatin (Byelorussian University, Minsk, Belarus), although various names have been proposed in the literature (see DeLoach et al., 2003). Ongoing research by our team (J.L. Tracy, ARS, Temple, TX; D.J. Kazmer, J.F. Gaskin, ARS, Sidney, MT; D.W. Bean, J.C. Herr, ARS, Albany, CA; A.A. Cossé, R.J. Bartelt, ARS, Peoria, IL; and D.C. Thompson, New Mexico State University, Las Cruces, NM) indicates the probability of four species involved in our studies.

The four populations of *D. elongata* included in this study originated from North Africa to western China. They were collected 15 km south of Sfax, Tunisia (latitude 34.66 N, longitude 10.67 E, elevation 10 m); 3 km west of Sfakaki, Crete, Greece (latitude 35.83 N, longitude 24.6 E, elevation 7 m); 7 km west of Karshi (Qarshi), Uzbekistan (latitude 38.86 N, longitude 65.72 E, elevation 350 m); and at the Turpan Eremophyte Botanic Garden of Academia Sinica, ca. 10 km southeast of Turpan, Xinjiang Province, China (latitude 42.86 N, longitude 89.22 E, elevation 70 m below sea level). We here refer to these as *D. elongata* from Tunisia, Crete, Uzbekistan, and Turpan. Although the first host specificity tests in 1992–1993 involved *D. elongata* from or near Turpan, survival on all test plants in the initial larval no-choice test was very low and no oviposition occurred in the adult test that included native *Frankenia* plants (DeLoach et al., 2003). Therefore, in the present study we evaluated this population more rigorously. In addition, several tests involving *D. elongata* from Crete included a comparison with the previously released population from Fukang, China. Voucher specimens of *D. elongata* from all locations were deposited with the National Collection of Insects and Mites of the National Museum of Natural History, Smithsonian Institution, Washington, DC (under Lot Numbers GSWRL-2004-02 and -2005-02).

All beetles from overseas (except from Fukang) were brought into the USDA-ARS, Exotic and Invasive Weed Research Unit quarantine facility at Albany, California where parasites, predators, and pathogens were eliminated. Beetles (eggs and/or adults) were subsequently sent to the USDA-ARS Arthropod Containment Facility (quarantine) at Temple, Texas to initiate our own colonies or for immediate use in some tests. We obtained Fukang beetles from field colonies near Lovell, Wyoming in June 2002 from our cooperator D.J. Kazmer (USDA-ARS, Sidney, Montana).