Prospects for the biological control of bellyache bush, *Jatropha gossypiifolia*

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Summary A project aimed at achieving biological control of bellyache bush, *Jatropha gossypiifolia*, has been in progress since 1997. A total of 177 sites in eight countries in tropical America and the Caribbean have been surveyed for natural enemies. A preliminary list of 60 species of phytophagous insects and one fungi have been identified from the plant. A database was designed to collate the information on the surveys for natural enemies. Seven species of insects and one fungus have been given further consideration. Four of these have been rejected because of a lack of sufficient host specificity or poor host compatibility. Three other insect species are currently being tested in quarantine. One of these, a seed-sucking bug appears to be suitable for release. Preliminary assessment of the rust fungus shows that it is a potentially feasible subject for further study. Details of the studies to arrive at the above conclusions, including foreign exploration, molecular taxonomy and host specificity testing are presented. Studies on the basic biology of the plant are providing an ecological foundation for the biological control project.

Keywords Biological control, tropical weeds, natural enemies.

INTRODUCTION

Bellyache bush, *Jatropha gossypiifolia* (Euphorbiaceae), is a serious emerging weed of Northern Australia. In the Weeds of National Significance prioritising exercise, bellyache bush was rated as number 21 (Thorpe and Lynch 2000). The Standing Committee on Agriculture and Resource Management (SCARM) has endorsed the weed as a target for biological control. It is native to the American tropics but has become widely naturalised elsewhere in the tropics where it has been introduced as an ornamental and medicinal plant. It invades rangelands particularly in riparian zones, but is also a serious problem in upland habitats, forming dense thickets which eliminate useful species, reduces productivity and hinders mustering. All parts of the plant especially the seeds are toxic and ingestion causes the deaths of grazing animals, a problem that occurs especially during drought.

Bellyache bush has been the target of considerable research activity in recent years and much progress has been made in the areas of ecology and management by herbicide, fire (work largely done by the Department of Natural Resources and Mines, Queensland) and biological control (largely by CSIRO). In this paper, we will focus on the efforts toward biological control.

MATERIALS AND METHODS

Surveys of the natural enemies have been conducted in the native range of this weed: tropical America and the Caribbean (Table 1). Surveys are one of the first steps in a biocontrol project and include the collection, rearing, curation, identification, cataloguing and literature searches of the natural enemies. A process of selecting the highest priority potential biocontrol agents was conducted. Permits were obtained from the Australian Quarantine and Inspection Service (AQIS) and Environment Australia (EA) to import into Australian quarantine many insect species for further testing.

RESULTS

Eight countries in the native range of this weed have been visited (Table 1). Searches have been made at a total of 177 locations. Some of these locations have been visited multiple times to generate 220 collection events. Thousands of insect specimens have been collected and sent for identification. A relational database using the program MS Access has been designed and all data has been entered for secure storage and querying. A preliminary list of phytophagous insects has been compiled consisting of 60 species in 21 families and four orders. One species of fungus has also been collected.

Table 1. Countries surveyed for natural enemies of *Jatropha gossypiifolia* and dates of surveys.

Country, State	Month/year
Mexico, Veracruz	95, 96, 97, 98, 99, 00, 01 (many dates)
Mexico, Pacific coast	11/95, 6/99, 7/99
Honduras	11/95, 6/99
Guatemala	6/99
Venezuela, west coast	10/97, 12/99, 2/00, 10/00
Venezuela, east coast	7/97, 10/97, 2/99
Dominican Republic	1/98, 9/98, 10-11/98
Puerto Rico	1/98
Trinidad	2/99
Curacao	12/99

The potential of these 60 insects as biological control agents is scarcely known. Fourteen species are known to have a wide host range and so may be dismissed. The host range of nearly all of the remaining 46 species was unknown.

Higher priority potential agents have been identified according to their abundance and the damage that they inflict in the native range. Permits have been obtained from AQIS and EA to import 10 insect species into Australian quarantine for further testing. Six of these have been imported for further assessment. One has been assessed in Mexico and another in the United Kingdom. The status of natural enemies that have been imported, tested or proposed as future agents is shown in Table 2.

Table 2. Status of candidate insects and pathogens studied for biological control of *Jatropha gossypiifolia*.

Agent	Part attacked	Schedule and status
Coleoptera: Cerambycidae		
Lagocheirus araneiformis	Stems	Imported and studied 98-00. Rejected.
Lagocheirus sp.	Stems	“
Styloleptus spp.	Stems	Imported twice 1998-99. Could not establish culture.
Leptostylus spp.	Stems	Permit for import obtained.
Lepturges spp.	Stems	“
Trachyderes spp.	Stems	“
Ozineus spp.	Stems	“
Parmenonta spp.	Stems	“
Hemiptera: Scutelleridae		
Pachycoris klugii	Fruits	Imported and studied 98-00. Rejected.
Agonosoma trilineatum	Seeds	Imported 2000. In culture. Specificity studies completed. Application for release pending.
Coleoptera: Chrysomelidae		
Colaspis sp. larvae – adults –	 Roots Leaves	Imported 2000. In culture for two generations. Some host testing commenced.
Uredinales		
Phakopsora jatrophicola	Leaves	Sent to CABI, UK 2000. Feasibility study completed
Lepidoptera: Tineidae		
Xylesthia sp.	Tips, leaves	To be imported 2002. Permit for import being prepared.
Lepidoptera: Noctuidae		
Spodoptera latifascia	Leaves	Tested 2001, Mexican Field Station. Not specific.
Coleoptera: Brentidae		
Gen. sp.	Young stems	To be imported 2002. Permit for import being prepared.

The most promising current agent so far is the seed feeding Scutelleridae *Agonosoma trilineatum.* Host testing in which newly emerged nymphs were placed on well developed fruits of test plant species has been applied to 74 species, each with four replicates. Development has not occurred on any species other than bellyache bush and other exotic *Jatropha* species. A second test of the response of adults to test plants has been conducted for a subset of 12 species. Adults did not survive long and did not oviposit, feed or mate on test plant species. Further information on the biology has been determined such as duration of the life cycle, fecundity and longevity of adults, and mode of feeding. The host test list for *Agonosoma trilineatum*, consisting of 49 species, 43 of which are in the Euphorbiaceae, was approved by AQIS and EA in June 2002. As this is the first plant species in the family Euphorbiaceae that has been target for biological control in Australia, challenges were experienced in compiling a host test list. We expect to submit an application for release of this insect in June 2002. The Tropical Weeds Research Centre (NRM Qld) is growing plants ready for the culturing of this insect following approval for release.

Lagocheirus spp. (Cerambycidae) beetles from Venezuela and Mexico were the subjects of intensive study in quarantine. In order to improve the success rate of rearing collected larvae to adults, we developed an artificial diet. The use of the diet also proved very effective for eliminating mites from the colonies. Once viable colonies were established, host testing commenced. Taxonomists told us that the individuals from Mexico and Venezuela were the same species, and furthermore that it is the pest of cassava, *Lagocheirus araneiformis*. However, we identified biological differences which led us to send specimens to the CSIRO Entomology molecular diagnostics laboratory in Canberra. They sequenced *ca.* 800 base pairs of the mitochondrial CO1 gene and *ca.* 600 base pairs of the nuclear ribosomal 28S gene. The results indicated that the taxa are indeed different species. Both species were then kept separate for further studies. A method of host testing *Lagocheirus* spp. was developed. The tests were sequential choice-minus control tests on combined oviposition and larval development. These beetles were eventually rejected as they feed on cassava, the important euphorbiaceous crop plant. The details of this interaction took a lot of time to unravel as feeding only occurred on cut stems of cassava and very rarely on living plants. However, since cassava is vegetatively propagated by stem cuttings, has potential as a crop in Australia and is currently very important crop in many parts of the world, a decision was made to cease testing and to eliminate colonies of these species.

Another promising agent is *Colaspis* sp. A culture was established in quarantine by breeding larvae on roots of potted plants. Unfortunately the colony did not thrive. Initial testing on adults feeding showed a high degree of specificity indicating promise for the future of this species if a method of rearing can be developed.

CABI BioScience, UK, conducted preliminary studies on the rust fungus, *Phakopsora jatrophicola*. We sent seeds and plants of bellyache bush which were established in the CABI glasshouse. We then sent the fungus from Mexico which was successfully reared in the lab. Although these preliminary results are positive, funds were not available to do a second stage study in which they would test the host specificity of the pathogen. Work will also be required to determine if the rust is autoecious or heteroecious. If heteroecious it may have other hosts in the sexual stage of its life cycle and testing would be more difficult. This initial work has provided an early indication of feasibility. We also need to test the effectiveness of the rust pathogen in the native range to determine whether expenditure on testing this fungus is justified.

Searching the literature on insect pests of the related plant, *Jatropha curcas* (a crop species in Central America) found potential new agents. A sap-sucking bug *Pachycoris klugii* (Scutelleridae) is recorded as the most damaging herbivore of plantations of *J. curcas* in Nicaragua and is also recorded as being specific to *Jatropha* spp. A permit was obtained to import *Pachycoris klugii* to determine whether it would develop on *J. gossypiifolia*. Shipments of eggs, nymphs and adults were made in 1999 from Chiapas State, Mexico. Unfortunately, this insect did not develop on *J. gossypiifolia* and the culture was destroyed. All other insect pests of *J. curcas* have been eliminated from consideration as biocontrol agents of *J. gossypiifolia* due to their unacceptably wide host range.

DISCUSSION

Several lines of evidence suggest that the prospects for biological control of bellyache bush are good. Firstly, there is a moderately large suite of natural enemies attacking this plant in the native range. Second, the plant is less vigorous in the native range and we attribute this to herbivory. Thirdly, no plant species in the tribe Jatropheae occur naturally or are economically important in Australia.

The size of the suite of natural enemies of bellyache bush (61 species) is not large compared to other tropical species such as mesquite (>945 species) (van Klinken and Campbell 2001); *Mimosa pigra* (c. 417 species) (Harley *et al.* 1995); four species of *Lantana* (550 species) (Palmer and Pullen 1995); *Parthenium hysterophorus* (262 species) (McClay *et al.* 1995); *Senna obtusifolia* (108 species) (Gillett *et al.* 1991), although it is comparable with *Sida acuta* (51 species) and *S. rhombifolia* (27 species) (Palmer and Pullen 2001). The results of these studies partially reflect sampling effort. Thus far, the effort on bellyache bush is not complete and we expect to find more organisms.

We expect that the fauna of a species of plant that is so well protected by toxic chemicals (Das and Das 1994) will be relatively small. We also expect however, that it may be a specialised fauna that is most host specific than that associated with a more benign plant. Within the plant, we expect that insects that feed on parts well protected by chemicals (e.g. young leaf and fruit feeders) are more specific than those feeding on parts less protected (e.g. wood borers, xylem suckers) (Schoonhoven *et al.* 1998, p. 51) and hence are better biological control prospects. We will test these hypotheses within this project.

Because no plant species in the tribe Jatropheae occur naturally or are economically important in Australia, the level of host specificity required of a biological control agent is not strict and the probability and consequences of non-target impact are minimal. The only other members of this tribe to occur in Australia are ornamental members of the *Jatropha* genus, none of which are economically important and several of which are weeds. The tribe Jatropheae occurs in the subfamily Crotonoideae with eight other tribes. Many of the approximately 90 members of the subfamily Crotonoideae present in Australia are valued plants. For example, the potential crop species cassava and native species of about 12 genera. These species are well represented on the host specificity test list.

The relational database proved very useful for storage and querying of native range data. Within the database, there are four primary 'categories' (tables) of information, storing data relating to:

- collection events (e.g. collection number, date),
- specimen information (e.g. insect identification number, number collected),
- locality information (e.g. locality name, country),
- insect information (e.g. taxonomy, geographic range).

As the database is relational, the tables are linked to one another through common fields. For example, the collection and the specimen tables are linked through the collection number, which is common to both tables. These links allow information to be extracted from different tables. For example, it is possible to identify all the localities from which a species was collected, even though the insect and locality tables are not directly linked to one another. The database can be used to rapidly generate useful information, including lists of insects associated with particular weeds, lists of collection sites and dates, and lists indicating when and where a particular insect species was collected. The database structure is being used for other weeds for which information is being collected on potential biological control agents.

The current geographic range of this plant is large because of its widespread distribution by humans for traditional medicine but the centre of origin is thought to be the drier islands of the Caribbean and the Venezuelan coastline. An analysis of the distribution of the insect species will be conducted to test this hypothesis of the location of the evolutionary centre. The diversity of natural enemies should be greater at this point.

This weed is also a serious problem in East Timor and east Indonesia. These countries would benefit from the extension of Australian research activities. Bellyache bush has been included in the CRC for Australian Weed Management as a case study of an invasive rangeland shrub. Future involvement of the CRC may include modelling the ecology and management and prioritising potential biocontrol agents. Ecologically, there are few close analogues to this weed. However, it is typical in the sense that it was an invasive weed occurring in low value country. The economics of attempting to control such a weed on extensive grazing systems applies to many weed syndromes such as mesquites, prickly acacia, rubber vine and parkinsonia.

Close cooperation between CSIRO and DNRM (Qld) and DBIRD (NT) has been a feature of this project. The biological control work described here has been conducted by CSIRO with funding input from DBIRD and DNRM. DNRM have simultaneously been working on other management options. The mass-rearing and release of the agents that have been approved for release will be done by DNRM and DBIRD. Evaluation of the impact of the agents will also be done by DNRM and has already commenced with baseline ecological parameters being collected for comparison with data after the agents are established and damaging plants. DBIRD has provided species of Euphorbiacae for host testing insects and cuttings of bellyache bush for insect cultures in Brisbane.

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