Exploration for potential biological control agents of *Euphorbia paralias*, a major environmental weed of coastal ecosystems in Australia

John K. Scott[1], Mireille Jourdan[2], Louise Morin[3], Thierry Thomann[2] and Paul B. Yeoh[1]
[1] CSIRO Entomology, Private Bag 5, PO Wembley, WA 6913, Australia
[2] CSIRO European Laboratory, Campus International de Baillarguet, 34980 Montferrier-sur-Lez, France
[3] CSIRO Entomology, GPO Box 1700, Canberra, ACT 2601, Australia
Corresponding author: John.K.Scott@csiro.au

Summary Surveys were conducted during 2008–2009 to identify herbivorous arthropods and plant pathogens associated with sea spurge (*Euphorbia paralias*) (Euphorbiaceae) in its introduced and native ranges. These surveys are preliminary to the initiation of a biological control program, which is currently the most sustainable option for long-term control of this weed of coastal dune systems in Australia. The survey within coastal sites in southern Australia did not identify any arthropods or pathogens specific to sea spurge. In contrast a rust fungus, other pathogens and insects with a host range potentially limited to sea spurge were located within its native range in coastal France. Potential options for biological control are reviewed here based on these findings.

Keywords Biological control, surveys for agents, insects, plant pathogens, France, Australia.

INTRODUCTION

Sea spurge (*Euphorbia paralias* L.) (Euphorbiaceae) is the major weed of the fore dune, and increasingly the hinterland, of coasts of southern Australia. It threatens a range of endangered species, aboriginal heritage sites and public recreational use of coasts. Sea spurge is being considered for nomination as a target for biological control, as this approach is the most viable option for long-term control.

This pilot project's main goal was to carry out preliminary surveys of natural enemies of sea spurge in its regions of origin, the Mediterranean coast and the Atlantic coast of Europe, to identify candidate biological control agents. Suitable candidates must be sufficiently host specific so that they will not have negative effects on non-target plants should they be released in Australia. In addition, initial surveys in Australia sought evidence for damaging insects or pathogens that may already be present so that resources are not wasted importing beneficial organisms that are already present.

This paper summarises results from the surveys. More details will be published separately on the results of the plant pathology investigations.

MATERIALS AND METHODS

Surveys in Australia Coastal sites in Western Australia (18), South Australia (3), Victoria (11) and Flinders Island/Tasmania (4) were examined between October 2008 and December 2009. Natural resource managers in these regions where sea spurge is a problem weed were asked to collect plants showing insect or pathogen damage.

Surveys in France Searches focused on sandy beaches with dunes. The Mediterranean coast was searched along 400 km from Cap de Sant Sebastià (northern Spain) to Sausset les Bains (near Marseille, France) during four surveys from July 2008 to July 2009. The Atlantic coast was searched along 200 km from Biarritz to Arcachon from 29 June to 1 July 2009.

Insects At each site overall levels of damage were visually assessed. Where possible, we collected 30 plants showing each kind of damage. When present, 100 seeds were collected, 20 were dissected and the remaining stored in gauzed plastic boxes and regularly checked to detect insect emergence.

Substantially damaged plant parts were dissected to locate possible causal organisms. All samples were placed separately in gauzed plastic boxes for insect emergence (one box per damage category per site). When young plants were collected with roots, they were planted into potting mixture in pots and placed within gauzed cages and watered until insect emergence. For at least 3 months the boxes and cages were regularly checked for insect emergence. The insects emerged were counted and identified.

Plant pathogens Prior to the surveys, records of fungi associated with sea spurge were searched in the literature to define the most promising agents.

At each site, plants were searched visually for evidence of disease symptoms. Samples of each category of symptoms were collected, placed in dry conditions between several layers of absorbent paper, pressed and brought back to the laboratory where they were examined. Fungi were isolated from each type of symptom, multiplied and tested for pathogenicity on sea spurge.

RESULTS

Surveys in Australia No arthropod or pathogen species with limited host ranges were identified from sea spurge in Australia. Likewise, no damage by gall-inducing or other specialised feeding damage was observed. No records of plant pathogens on sea spurge in Australia were found in the Australian Plant Pest Database.

No additional reports from surveying resource managers were received of sea spurge showing damage from host-specific pathogens (especially rusts) or insects, although some minor damage (black spots) of unknown cause was reported.

Surveys in France Sea spurge with evidence of damage was sampled at 14 sites on the Mediterranean coast and 8 sites on the Atlantic coast out of the 39 sites surveyed. Sea spurge was found on stable dunes. It was rare from Spain to Perpignan due to intensive tourism activities and the difficulty of access to coastal regions where natural vegetation occurs. It was more abundant and accessible along the French coast from Perpignan to Sausset les Bains and on the west coast.

Insects Insects from four families (Table 1) included two Lepidoptera and one Diptera species damaging stems and one Coleoptera species mining roots and stems. No insects emerged from fruits or seeds. No insect or fungal damage was found in the 20 seeds dissected per sample.

Acroclita subsequana This insect was widespread on sea spurge on Mediterranean and Atlantic coast during the surveys and its larvae were responsible for the main damage observed. The larvae tie together leaves, apex of stems or inflorescence causing dehiscence of stems. By damaging inflorescences and fruits they affect the reproductive success of plants. According to Razowski (2003) and our observations, *A. subsequana* has two generations per year (one in spring, one in autumn). The pupa hibernates. Sobhian (1996) also collected *A. subsequana* from *Euphorbia characias* L. in southern France, but found that field-collected insects would not feed on *Euphorbia esula* L., a target for biological control in USA, whereas neonate larvae would.

According to Razowski (2003) the genus *Acroclita* has eight species of Palaearctic and European origins and host plants are in the Euphorbiaceae. Horak (2006), however, has the *Acroclita* and related genera present in Australia and Asia, where they are recorded on a wide range of hosts (but not *Euphorbia*). The host range of *A. subsequana* cannot be determined from the limited information published on this species, although the Razowski (2003) review of tortricids of Europe lists sea spurge as the sole host. This, and the level of damage caused to sea spurge, indicates that this insect should be investigated further as a candidate biological control agent.

Eucrostes indigenata This species was found during our surveys at only one site and its damage was similar to that of *A. subsequana*. It is also bivoltine or often trivoltine (Hausmann 2001). Hausmann (2001) states that *E. indigenata* is monophagous on *Euphorbia* and is found in coastal habitats. Further investigation is required to fully assess its potential for biological control.

Liriomyza pascuum We found larvae of this species mining leaves of sea spurge at only some sites. The insect damage was found when leaves were examined with a binocular microscope in the laboratory and not in the field. This means that this insect could be more widespread than was recorded. This species is common on other *Euphorbia* species. (Martinez and Sohbian 1998), but its host range is unknown as is the impact of leaf mining on the biology of the plant.

Mordellistena pseudopumila This insect is an opportunist feeder, probably on decaying stem material (cf. Ford and Jackman 1996), not host-specific to the genus *Euphorbia* and thus unsuitable as a potential biological control agent.

Plant pathogens Six fungi were found associated with diseased sea spurge. Five were provisionally identified to the genus level and one remains uncertain (Table 2). Three of them (*Melampsora euphorbiae*, the unidentified pathogen and *Phomopsis euphorbiae*) were pathogenic to sea spurge.

Melampsora euphorbiae This rust fungus is considered as a good candidate biological control agent because it does not have an alternate host and its host range is restricted to a few species of *Euphorbia* (Bruckart *et al.* 1986). Specificity of *M. euphorbiae* was previously reported by Müller (1907) and Jorstad (1954) who recognised it as a species complex consisting of several biological races or special forms, each infecting only one or two species of *Euphorbia*. Indeed, *M. euphorbiae* is reported as a pathogen of more than 50 species of *Euphorbia*, including *Euphorbia peplus* L. in Australia (Farr and Rossman 2010).

Table 1. Insect species and number of sites with associated damage detected during the surveys of 22 sites.

Family	Insect species	Fruits	Flower stems	Stems	Stem tips	Stem mines
Agromyzidae	*Liriomyza pascuum* (Meigen)			1		
Geometridae	*Eucrostes indigenata* (Villers)			1		
Mordellidae	*Mordellistena pseudopumila* (Ermisch)					8
Tortricidae	*Acroclita subsequana* (Herrich-Schäffer)	7	13	14	3	

Table 2. Fungal species reported on sea spurge in Farr and Rossman (2010) plus species detected during the surveys of 22 sites.

Class or Family	Fungi	Recorded distribution[A]	No. of sites with symptoms	Pathogenic on sea spurge
	Unidentified pathogen		9	Yes
Coelomycetes	*Macrophoma* sp.		4	No
Coelomycetes	*Phomopsis euphorbiae* (Sacc.) Traverso	Greece	3	Yes
Hyphomycetes	*Alternaria* sp.		10	No
Hyphomycetes	*Stemphylium* sp.		12	No
Hyponectriaceae	*Physalospora euphorbiae* (Plowr. & Phill.) Sacc.	Spain	0	
Melampsoraceae	*Melampsora euphorbiae* (Schub.) Castagne or *Melampsora* sp.	Bulgaria, Greece, UK, Libya, Morocco, Libya[B]	13	Yes
Mycosphaerellaceae	*Mycosphaerella euphorbiae* Niessl ex J. Schröt.	UK	0	
Peronosporaceae	*Peronospora valesiaca* Gäumann	UK	0	
Pleosporaceae	*Pleospora herbarum* (Pers.) Rabenh.	Greece	0	
Pleosporaceae	*Pyrenophora euphorbiae* Politis	France	0	

[A] Presence of distribution records indicates species recorded in Farr and Rossman (2010). Data of the distribution on sea spurge only.
[B] As the synonym *Melampsora helioscopiae*.

Unidentified fungus The damage caused by this fungus in the field and during preliminary pathology studies was impressive. More information on its identity (using traditional and molecular techniques) is needed, as well as preliminary host-range tests, to assess its potential as a biological control agent. A correct identification for this fungus would also enable us to check if it has been recorded in Australia by consulting mycological herbaria and literature records.

Phomopsis euphorbiae This fungus was also damaging towards a French sea spurge accession during pathogenicity tests, but additional tests on Australian plant accessions and other key non-target species are required to assess its biological control potential.

DISCUSSION

Four insect species were found on sea spurge in its native habitat in France, three of which could be suitable for use in biological control. These were two moths with larvae that feed on leaf stems and inflorescence, the tortricid *A. subsequana* and the geometrid *E. indigenata,* and an agromyzid leaf mining fly, *Liriomyza pascuum*. The rust, *M. euphorbiae*, and an unidentified fungus that is very damaging to plants were also found. Both fungi were pathogenic towards Australian sea spurge accessions, a key prerequisite for their continued investigation as potential biological control agents.

The area surveyed in the native range was a relatively small part of the natural distribution of sea spurge and a wider survey will probably produce further potential agents. Very few species are currently known from sea spurge, possibly because its specialised littoral habitat may be hostile to many herbivores. However, sea spurge has never been surveyed before now despite the considerable survey activity on related species that are targets for biological control in North America.

Arthropods likely to be encountered in further surveys include *Aphthona flaviceps* Allard (Coleoptera: Chrysomelidae). The genus is associated with *Euphorbia* species and the larvae feed on the roots (Doguet 1994). Species within the *Hyles euphorbiae* complex (Lepidoptera: Sphingidae) (Hundsdoerfer *et al.* 2005) with large leaf feeding larvae may be found on both sides of the Mediterranean. In a third example, Graham (1984) describes seed feeding wasps, *Erytoma* species, which may be found on sea spurge.

Priorities for future research were identified including host range studies and molecular identification (including phylogenies) for all species with biological control potential. The highest priorities, due to the likelihood of a very high level of host specificity and potential for significant damage, are to examine further the host range of the sea spurge strain of the rust *M. euphorbiae* and the tortricid, *A. subsequana*.

ACKNOWLEDGMENTS

We thank Mic Neave, Don Gomez, Noboru Ota, Sauna Potter and Dr Roger Shivas for assistance in Australia. We gratefully acknowledge the funding support of CSIRO and the Tasmanian Department of Primary Industries and Water. The authors also thank Michel Martinez (Agromyizidae and Tachinidae), Christian Cocquempot (Tortricidae and Geometridae) and Pascal Leblanc (Mordellidae) for their fast and accurate insect identification and René Sforza and Janine Vitou for their help in France. We also thank Tim Heard and Mic Julien for their comments on the manuscript.

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