

# Control of freshwater invasive species

Global evidence for the effects of selected interventions



**David C. Aldridge, Stephanie L. Aldridge, Angela Mead, Nancy Ockendon, Ricardo Rocha, Helen Scales, Rebecca K. Smith, Alexandra Zieritz and William J. Sutherland**

**SYNOPSIS OF CONSERVATION EVIDENCE SERIES**

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David C. Aldridge, Stephanie L. Aldridge, Angela Mead, Nancy  
Ockendon, Ricardo Rocha, Helen Scales, Rebecca K. Smith, Alexandra  
Zieritz and William J. Sutherland

Synopses of Conservation Evidence

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## Advisory Board

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## About the authors

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**David Aldridge** is a Senior Lecturer in the Department of Zoology at the University of Cambridge and is Managing Director of Cambridge Environmental Consulting, UK.

**Stephanie Aldridge** is a Director of Cambridge Environmental Consulting, UK.

**Angela Mead** is a researcher for Cambridge Environmental Consulting, UK.

**Nancy Ockendon** is a post-doctoral researcher in the Department of Zoology, University of Cambridge, UK.

**Ricardo Rocha** is a post-doctoral researcher in the Department of Zoology, University of Cambridge, UK.

**Helen Scales** is an aquatic ecologist, author and researcher for Cambridge Environmental Consulting, UK.

**Rebecca Smith** is a Senior Research Associate in the Department of Zoology, University of Cambridge, UK.

**Alexandra Zieritz** is a post-doctoral researcher at the University of Nottingham, Malaysia Campus, Malaysia.

**William Sutherland** is the Rothschild Professor of Conservation Biology in the Department of Zoology, University of Cambridge, UK.

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# 1 About this synopsis

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## The purpose of Conservation Evidence synopses

Conservation Evidence synopses <b>do</b>	Conservation Evidence synopses <b>do not</b>
<ul style="list-style-type: none"><li>• Bring together scientific evidence captured by the Conservation Evidence project (over 5,400 studies so far) on the effects of interventions to conserve biodiversity</li></ul>	<ul style="list-style-type: none"><li>• Include evidence on the basic ecology of species or habitats, or threats to them</li></ul>
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<ul style="list-style-type: none"><li>• Work in partnership with conservation practitioners, policymakers and scientists to develop the list of interventions and ensure we have covered the most important literature</li></ul>	<ul style="list-style-type: none"><li>• Provide recommendations for conservation problems, but instead provide scientific information to help with decision-making</li></ul>

## Who is this synopsis for?

If you are reading this, we hope you are someone who has to make decisions about how best to support or conserve biodiversity. You might be a land manager, a conservationist in the public or private sector, a farmer, a campaigner, an advisor or consultant, a policymaker, a researcher or someone taking action to protect your own local wildlife. Our synopses summarize scientific evidence relevant to your conservation objectives and the actions you could take to achieve them.

We do not aim to make your decisions for you, but to support your decision-making by telling you what evidence there is (or isn't) about the effects that your planned actions could have.

When decisions have to be made with particularly important consequences, we recommend carrying out a systematic review, as the latter is likely to be more comprehensive than the summary of evidence presented here. Guidance on how to carry out systematic reviews can be found from the Centre for Evidence-Based Conservation at the University of Bangor ([www.cebc.bangor.ac.uk](http://www.cebc.bangor.ac.uk)).

## The Conservation Evidence project

The Conservation Evidence project has four parts:

1) An online, **open access journal** *Conservation Evidence* that publishes new pieces of research on the effects of conservation management interventions. All our papers are written by, or in conjunction with, those who carried out the conservation work and include some monitoring of its effects.

2) An ever-expanding **database of summaries** of previously published scientific papers, reports, reviews or systematic reviews that document the effects of interventions.

3) **Synopses** of the evidence captured in parts one and two on particular species groups or habitats. Synopses bring together the evidence for each possible intervention. They are freely available online and many are available to purchase in printed book form.

4) **What Works in Conservation** this publication includes key results from the collated evidence for each intervention for each species group or habitat, along with an assessment of its effectiveness by expert panels.

These resources currently comprise over 5,400 pieces of evidence, all available in a searchable database on the website [www.conservationevidence.com](http://www.conservationevidence.com).

Alongside this project, the Centre for Evidence-Based Conservation ([www.cebc.bangor.ac.uk](http://www.cebc.bangor.ac.uk)) and the Collaboration for Environmental Evidence ([www.environmentalevidence.org](http://www.environmentalevidence.org)) carry out and compile systematic reviews of evidence on the effectiveness of particular conservation interventions. These systematic reviews are included on the Conservation Evidence database.

## Scope of the Freshwater Invasive Species synopsis

Nonindigenous species constitute a growing concern to environmental managers and stakeholders because of their broad-ranging impacts on biodiversity, ecosystem services and economies (Gallardo & Aldridge 2013). In Europe, approximately 20% of the 12,000 non-native species described have been already reported in Great Britain (Gallardo *et al.* 2013), with total annual costs for the British economy estimated at £1.7 billion (Williams *et al.* 2010).

Freshwater ecosystems have been especially affected by nonindigenous species (Sala *et al.* 2000; Leprieur *et al.* 2008). Species have been introduced through intentional vectors such as stocking of lakes and rivers with sport and food organisms (Gido & Brown 1999), the trade in ornamental plants and animals for ponds and aquaria (Keller & Lodge 2007), and aquaculture (Holdich *et al.* 1999). Freshwater species have also been introduced unintentionally, such as through ballast water discharge (Ricciardi 2006) and as contaminants of aquarium plants (Keller & Lodge 2007).

Great Britain has a long history of international trade, and many nonindigenous freshwater species have established widely (Keller *et al.* 2009). For example, the zebra mussel *Dreissena polymorpha* was introduced as a contaminant of imported timber in the 19<sup>th</sup> Century, and now is widespread through much of England (Aldridge 2010). Zebra mussels can drive ecological change, cause declines in native organisms (Aldridge *et al.* 2004) and cause a fouling nuisance in raw water pipelines

(Elliott *et al.* 2005) which has resulted in an annual management costs in Britain of approximately £5 million (Oreska & Aldridge 2010). Crayfish plague *Aphanomyces astaci* was introduced into Great Britain in the 1970s when infected North American crayfish were introduced for aquaculture purposes. The plague threatens Britain's one native crayfish *Austroptamobius pallipes* which is declining rapidly as a consequence. A number of nonindigenous aquatic plants have also established in Great Britain, including the New Zealand pigmyweed *Crassula helmsii*, which was imported by the nursery trade in the 20<sup>th</sup> century. Like many non-native plants, this species can form a monoculture which excludes native plants, impedes water flow and affects recreational activities (Dawson & Warman 1987).

In recent years a particularly important donor 'hot spot' for freshwater nonindigenous species into Western Europe and Great Britain has been the Ponto-Caspian region, located between the Black, Azov and Caspian Seas (Bollache *et al.* 2008). Over one hundred species are known to have spread from this region (e.g. Alexandrov *et al.* 2007). The spread has been facilitated by the construction of canals which link together once isolated river systems (bij de Vaate *et al.* 2002). The successful establishment of so many of these species is likely to reflect the similar climates found between Western Europe and the Ponto-Caspian (Gallardo & Aldridge 2013) and may also result from positive interactions between the invaders which consequently facilitates their spread (known as 'invasional meltdown').

Recent horizon-scanning activities in Great Britain (e.g. Gallardo & Aldridge 2013; Roy *et al.* 2014) have helped to identify lists of priority freshwater taxa which either have the potential to establish and spread in Great Britain, or which have established locally in Britain and are predicted to become widespread and damaging. In order for Great Britain to respond quickly and appropriately to the discovery of a high-risk invader, rapid response plans are needed. Selection of the most appropriate control and management methods require a sound evidence base on their efficacy and suitability for use within particular scenarios. The importance of having a sound evidence base for management interventions triggered the initiation of the Conservation Evidence ([www.conservationevidence.com](http://www.conservationevidence.com)) project. This synopsis builds on the Conservation Evidence methodology to provide a directory of evidence-based interventions for managing freshwater taxa which are considered of high risk to Great Britain's ecosystems or economy. The synopsis can help managers reach fast and informed decisions on the most appropriate response to a newly discovered invader.

This synopsis covers evidence for the effects of interventions to enhance the management and control of freshwater invasive species in the UK. Any bias towards evidence from particular countries reflects a current bias in the published research available. The synopsis does not include evidence from the substantial literature on the interactions between invasive species, local biodiversity and freshwater environments.

'Freshwater' was defined as any species that requires being at least partly submersed in freshwater to complete its life cycle or that is regularly sampled from freshwaters. For most species (e.g., fishes and macrophytes) this distinction is clear. Others, particularly wetland plants, are less easy to classify, and in such cases, we deferred to categorisations published by others.

It is important to note that many management responses to nonindigenous species may require specific regulatory approvals to be granted before they can be carried out. Consideration must be made not only of the species to be controlled, but also of the impacts of the intervention on non-target organisms, the wider ecosystem and on other uses of the waterbody. Regulations change continually, and so it is essential that relevant authorities are consulted before any interventions are undertaken. In addition, selection of the most suitable intervention must take into account its wider acceptability to stakeholders and the general public, a balance between the cost and likely benefits, and an assessment of the likelihood of reinvasion.

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- Ricciardi, A. (2006) Patterns of invasion in the Laurentian Great Lakes in relation to changes in vector activity. *Diversity and Distributions*, 12, 425–433.
- Roy, H.E., Peyton, J., Aldridge, D.C. et al. (2014) Horizon scanning for invasive alien species with the potential to threaten biodiversity in Great Britain. *Global Change Biology*, 20, 3859–3871.
- Sala, O.E., Chaplin, F.S, Armesto, J.J. et al. (2000) Biodiversity—global biodiversity scenarios for the year 2100. *Science*, 287, 1770–1774.
- Williams, F., Eschen, R., Harris, A., Djeddour, D., Pratt, C., Shaw, R.S. & Murphy, S.T. (2010) The economic cost of invasive non-native species on Great Britain. CABI report, 198pp.

## How we decided which conservation interventions to include

In order to achieve the maximum degree of relevance and usefulness to practitioners, a priority list of freshwater invasive species was developed, as identified by the Advisory Board. The Advisory Board included key representatives of UK governmental agencies with a lead role in the management of freshwater invasive species. Firstly, a priority long-list was produced by selecting all freshwater species featured in one or more of the following eight previously published priority or alert lists:

1. GB Non-native Species Secretariat Alert species, available at <https://secure.fera.defra.gov.uk/nonnativespecies/factsheet/index.cfm>,
2. Environment Agency top 10, available at [http://www.environment-agency.gov.uk/static/documents/Invasive\\_species\\_top\\_10\\_hit\\_list.pdf](http://www.environment-agency.gov.uk/static/documents/Invasive_species_top_10_hit_list.pdf),
3. Parrott *et al.* (2009), available at <http://publications.naturalengland.org.uk/publication/43005>,
4. DAISIE 100 worst, available at <http://www.europe-aliens.org/speciesTheWorst.do>,
5. ISSG 100 worst, available at <http://www.issg.org/publications.htm#worst100>,
6. SEBI 2010, available at <https://wcd.coe.int/com.instranet.InstraServlet?command=com.instranet.CmdBlobGet&InstranetImage=1298206&SecMode=1&DocId=1438902&Usage=2>
7. GB Non-native Species Secretariat shortlist of Ponto-Caspian Amphipoda, unpublished.
8. UK Water Framework Directive priority list, available at <http://www.wfduk.org/tagged/alien-species>

The resulting priority long-list spanned 172 species. Of these 172 species, 10 species or taxonomic groups were identified by the Advisory Board in priority order. It was decided that the priority list required a thorough review of the literature. Selection was based on the following criteria: (1) there was a perceived ecological and/or economic risk or impact to the UK; (2) there was a current lack of knowledge on management and control options for the species; (3) the species was not already being reviewed in detail in other projects (for this reason, the quagga mussel *Dreissena rostriformis bugensis* was not included on this list); and (4) the final list included broad taxonomic coverage. This is because management tools are likely to be translatable to similar groups of organisms. The species and taxonomic groups, in priority order are as follows:

### A – Highest Priority

1. Arthropoda (Crustacea): Ponto-Caspian gammarids
2. Chordata (Fish): Ponto-Caspian gobies
3. Arthropoda (Crustacea): Crayfish *Procambrus* spp.
4. Plants: Water-primrose *Ludwigia* spp.
5. Chordata (Fish): Black *Ameiurus melas* and brown *A. nebulosus* bullhead

### B - Mid Priority

6. Mollusca (Bivalvia): Asian clams *Corbicula fluminalis*
- C – Lower priority
7. Chordata (Amphibian): American bullfrog *Lithobates catesbeianus*
  8. Plants: Skunk cabbage *Lysichiton americanus*
  9. Plants: Floating pennywort *Hydrocotyle ranunculoides*
  10. Chordata (Reptiles): Red-eared terrapin *Trachymes scripta*

We are continuing to add new species and taxonomic groups to this synopsis, and the invasive plants, New Zealand pigmyweed *Crassula helmsii* and Parrot's feather *Myriophyllum aquaticum*, have now been added. These species were prioritised in response to a desire for information about these species from Conservation Evidence users.

### How we reviewed the literature

In addition to evidence already captured by the Conservation Evidence project, we have searched the following sources for evidence relating to control of freshwater invasive species:

- Four specialist journals, from their first publication to the end of April 2013 (*Aquatic Invasions*, *Aquatic Conservation: Marine and Freshwater Ecosystems*, *Biological Invasions* and *Invasive Plant Science and Management*; each journal was systematically screened from first date of publication, 2006, 1991, 1999 and 2008 respectively to the latest volume/issue available up until April 2013, volume 8, issue 1, volume 23, issue 2, volume 15, issue 4 and volume 9, issue 3 respectively). For parrot's feather the search was extended to 23 additional specialist journals (*Applied Vegetation Science*, *Aquatic Botany*, *Aquatic Ecology*, *Aquatic Ecosystem Health & Management*, *Aquatic Living Resources*, *Biocontrol*, *Biocontrol Science and Technology*, *Biological Control*, *Freshwater Biology*, *Freshwater Science*, *Hydrobiologia*, *Journal of Aquatic Plant Management*, *Journal of Great Lakes Research*, *Journal of Vegetation Science*, *Limnologica*, *Management of Biological Invasions*, *NeoBiota*, *Weed Biology and Management*, *Weed Research*, *Wetlands*, *Wetlands Ecology and Management*), which were screened from the first date of publication up until December 2016.
- Thirty general conservation journals over the same time period.

The criteria for inclusion of studies in the Conservation Evidence database are as follows:

- There must have been an intervention carried out that conservationists would do.
- The effects of the intervention must have been monitored quantitatively.

These criteria exclude studies examining the effects of specific interventions without actually doing them. For example, predictive modelling studies and studies looking at species distributions in areas with long-standing management histories (correlative studies) were excluded. Such studies can suggest that an intervention

could be effective, but do not provide direct evidence of a causal relationship between the intervention and the observed biodiversity pattern. Any action that aimed at managing or controlling an invasive freshwater species was considered a relevant intervention. This included actions towards preventing the arrival or spread of invasive species, but did not include detection or monitoring.

Additional targeted searches were conducted using the ISI Web of Knowledge and a set of key search terms. A trial search was undertaken using the quagga mussel as the subject. It was decided that the scientific name of the species involved should be used rather than the common name (“Dreissena + bugensis” yielded 501 manuscripts, compared with 387 for “quagga mussel”). Of the 501 manuscripts, a manual search of the titles revealed 29 manuscripts that were likely to be relevant. In comparison, searching through each of the 501 manuscripts using the following search terms: “Dreissena + bugensis + control” and “Dreissena + bugensis + management” 19 and 17 relevant manuscripts were identified (seven of which had not been identified using title alone). The search terms also reduced the manuscript numbers to a more manageable level for more intense screening. Therefore the following was agreed with the Advisory Board. For each priority species other than parrot’s feather:

1. Search the species scientific name. (Note: if fewer than 100 papers are listed, then all can be screened by title/abstract).
2. If more than 100 papers are listed, refine the search by adding the terms “control” and “management”. Screen title/abstract of all remaining manuscripts.

For parrot’s feather the ISI Web of Knowledge search using the scientific name (“Myriophyllum + aquaticum”) yielded 187 manuscripts, all of which were screened.

The wider grey literature was identified with guidance from the expert Advisory Board and through approaching additional experts researching these species and taxonomic groups.

### **How the evidence is summarized**

In the text of each section, studies are presented in chronological order, so the most recent evidence is presented at the end. The summary text at the start of each section groups studies according to their findings.

At the start of each chapter, a series of **key messages** provides a rapid overview of the evidence. These messages are condensed from the summary text for each intervention.

Background information is provided where we feel recent knowledge is required to interpret the evidence. This is presented separately and relevant references included in the reference list at the end of each background section.

The information in this synopsis is available in two ways:

- As a pdf to download from [www.conservationevidence.com](http://www.conservationevidence.com).
- As text for individual interventions on the searchable database at [www.conservationevidence.com](http://www.conservationevidence.com).

### Terminology used to describe evidence

Unlike systematic reviews of particular conservation questions, we do not quantitatively assess the evidence or weight it according to quality. However, to allow you to interpret evidence, we make the size and design of each trial we report clear. The table below defines the terms that we have used to do this.

The strongest evidence comes from randomized, replicated, controlled trials with paired-sites and before and after monitoring.

<b>Term</b>	<b>Meaning</b>
<b>Site comparison</b>	A study that considers the effects of interventions by comparing sites that have historically had different interventions or levels of intervention.
<b>Replicated</b>	The intervention was repeated on more than one individual or site. In conservation and ecology, the number of replicates is much smaller than it would be for medical trials (when thousands of individuals are often tested). If the replicates are sites, pragmatism dictates that between five and ten replicates is a reasonable amount of replication, although more would be preferable. We provide the number of replicates wherever possible, and describe a replicated trial as 'small' if the number of replicates is small relative to similar studies of its kind. In the case of translocations or release of animals, replicates should be sites, not individuals.
<b>Controlled</b>	Individuals or sites treated with the intervention are compared with control individuals or sites not treated with the intervention.
<b>Paired sites</b>	Sites are considered in pairs, when one was treated with the intervention and the other was not. Pairs of sites are selected with similar environmental conditions, such as soil type or surrounding landscape. This approach aims to reduce environmental variation and make it easier to detect a true effect of the intervention.
<b>Randomized</b>	The intervention was allocated randomly to individuals or sites. This means that the initial condition of those given the intervention is less likely to bias the outcome.
<b>Before-and-after trial</b>	Monitoring of effects was carried out before and after the intervention was imposed.

<b>Review</b>	A conventional review of literature. Generally, these have not used an agreed search protocol or quantitative assessments of the evidence.
<b>Systematic review</b>	A systematic review follows an agreed set of methods for identifying studies and carrying out a formal 'meta-analysis'. It will weight or evaluate studies according to the strength of evidence they offer, based on the size of each study and the rigour of its design. All environmental systematic reviews are available at: <a href="http://www.environmentalevidence.org/index.htm">www.environmentalevidence.org/index.htm</a>
<b>Study</b>	If none of the above apply, for example a study looking at the number of people that were engaged in an awareness raising project.

### **Significant results**

Throughout the synopsis we have quoted results from papers. Unless specifically stated, these results reflect statistical tests performed on the results.

### **Multiple interventions**

Some studies investigated several interventions at once. When the effects of different interventions are separated, then the results are discussed separately in the relevant sections. However, often the effects of multiple interventions cannot be separated. When this is the case, the study is included in the section on each intervention, but the fact that several interventions were used is made clear.

# 1 Invasive plants

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## 1.1 Floating pennywort *Hydrocotyle ranunculoides*

### Background

In its native range, floating pennywort *Hydrocotyle ranunculoides* occurs in, and at the margins of, slowly flowing, warm and nutrient rich water in Argentina, Brazil and Paraguay, also in southern states of the USA. Floating pennywort is an invasive aquatic weed in North Western Europe and several other countries worldwide, including Chile, Australia and Uganda. In Europe, it is found in and around canals, lakes, rivers, streams, ditches, and garden ponds (Plant Protection Service *et al.* 2011).

Floating pennywort grows rooted in the mud along canal and pond margins, and out onto the water as a floating mat (Cordo *et al.* 1982). Rafts reduce light levels penetrating the water column and subsequent die-off of underlying plants and algae can lead to deoxygenation. This can result in fish mortality and changes to the invertebrate community (Stiers *et al.* 2009). The plant can also block waterways, which impedes navigation, prevents angling access and increases the risk of flooding.

Outside of its native range, floating pennywort competes with many native plant species. These include littoral marsh plants as well as submerged aquatic plants. These are overgrown and shaded out by the extensive beds or floating carpets (Cordo *et al.* 1982). Species richness of native aquatic plants may be reduced by more than 50% and submerged species may disappear entirely (Nijs *et al.* 2009).

Vegetative reproduction is the most common way of dispersion, and takes the form of stem fragments detaching from parent mats to spread downstream and colonize further sites. Regeneration capacity of floating pennywort is high since a new plant can be formed from a single node fragment (Hussner & Lössch 2007). There are indications that flowering and seed production are stimulated by conditions unfavourable for vegetative growth (Baas & Duistermaat 1999).

Floating pennywort is difficult to detect at an early stage of invasion, and therefore control or eradication actions often start when the plant is already well-established.

Baas W.J. & Duistermaat L.H. (1999) The invasion of floating pennywort (*Hydrocotyle ranunculoides* Lf) in the Netherlands, 1996-1998. *Gorteria*, 25, 77-82.

Cordo, H. A., DeLoach C. J. & Ferrer R. (1982) The weevils *Lixellus*, *Tanysphiroideus*, and *Cyrtobagous* that feed on *Hydrocotyle* and *Salvinia* in Argentina. *Coleopterists Bulletin*, 36, 279-286.

Hussner, A. & Lössch, R. (2007) Growth and photosynthesis of *Hydrocotyle ranunculoides* L. fil. in Central Europe. *Flora (Jena)*, 202, 653-660.

Nijs, I., Verlinden, M., Meerts, P., Dassonville, N., Domken, S., Triest, L., Stiers, I., Mahy, G., Saad, L., Lebrun, L., Jacquemart, A.L. & Cawoy, V. (2009) Biodiversity impacts of highly invasive alien

plants: mechanisms, enhancing factors and risk assessment – Alien Impact. *Final report phase 1, BELSPO contract number SD/BD/01A*, Brussels, 50pp.

Plant Protection Service, Plant Research International Wageningen UR, Aquatic Ecology and Water Quality Management Group & Centre for Ecology and Hydrology Wallingford (2011) *EUPHRESKO DeCLAIM Final report A State of the art June 2011. Hydrocotyle ranunculoides* L.f. 69pp.

Stiers, I., Crohain, N., Josens, G. & Triest, L. (2009) Impact of aquatic invasive species on native plant and benthic invertebrate assemblages in Belgian ponds, in: Pieterse, A., Rytönen, A.M. & Hellsten, S. (eds) *Aquatic weeds 2009. Reports of Finnish Environment Institute*, 15, 120-121.

## **Key messages**

### ***Biological control using co-evolved, host-specific herbivores***

A replicated laboratory and field study in South America found that the South American weevil fed on water pennywort but did not reduce the biomass.

### ***Biological control using native herbivores***

No evidence was captured on biological control of floating pennywort using native herbivores.

### ***Biological control using fungal-based herbicides***

No evidence was captured on the biological control of floating pennywort using fungal-based herbicides.

### ***Physical removal***

Two studies, one in Western Australia and one in the UK, found physical removal did not completely eradicate floating pennywort.

### ***Chemical control using herbicides***

A controlled, replicated field study in the UK found that the herbicide 2,4-D amine achieved almost 100% mortality of floating pennywort, compared with the herbicide glyphosate (applied without an adjuvant) which achieved negligible mortality.

### ***Combination treatment using herbicides and physical removal***

A before-and-after study in Western Australia found that a combination of cutting followed by a glyphosate chemical treatment, removed floating pennywort.

### ***Use of hydrogen peroxide***

A controlled, replicated study in the Netherlands found that hydrogen peroxide sprayed on potted floating pennywort plants at 30% concentration resulted in curling and transparency of the leaves but did not kill the plants.

### ***Use of liquid nitrogen***

No evidence was captured on the use of liquid nitrogen for control of floating pennywort.

### ***Flame treatment***

A controlled, replicated study in the Netherlands found that floating pennywort plants were killed by a three second flame treatment with a three second repeat treatment 11 days later.

### ***Excavation of banks***

No evidence was captured on the effects of excavation of banks using a sod-cutter or 'turf-cutter' to remove floating pennywort.

***Environmental control (e.g. shading, increased flow, reduction of rooting depth, or dredging)***

No evidence was captured on the potential for environmental control of floating pennywort using shading, increased flow, reduction of rooting depth to below one metre, or dredging.

***Public education***

No evidence was captured on the impact of education programmes on control of floating pennywort.

**1.1.1 Biological control using co-evolved, host specific herbivores**

- A replicated laboratory and field study in South America<sup>1</sup> found that the South American weevil caused more feeding lesions on floating pennywort than on any other plant species, but field results found that the weevil did not reduce floating pennywort biomass.

**Background**

One-off introduction of a co-evolved, host-specific species from the area of origin of the invasive pest can potentially provide sustainable control without affecting non-target native plants. For example, a laboratory study in Argentina, found that the South American weevil *Listronotus elongatus* appears host specific on floating pennywort *Hydrocotyle ranunculoides*, exhibiting both feeding and reproductive behaviour (Cordo *et al.* 1982). Observations have also been reported of extensive damage caused by this same weevil to floating pennywort (Newman & Duenas 2010). In addition, observations in Germany showed that the introduced Coypus *Myocastor coypus* can eat floating pennywort (Hussner & Lösch 2007).

Cordo, H. A., DeLoach C. J. & Ferrer R. (1982) The weevils *Lixellus*, *Tanysphiroideus*, and *Cyrtobagous* that feed on *Hydrocotyle* and *Salvinia* in Argentina. *Coleopterists Bulletin*, 36, 279–286.

Hussner, A. & Lösch, R. (2007) Growth and photosynthesis of *Hydrocotyle ranunculoides* L. fil. in Central Europe. *Flora (Jena)*, 202, 653-660.

Newman J.R. & Duenas M.A. (2010) Information sheet: control of floating pennywort (*Hydrocotyle ranunculoides*). Centre for Ecology & Hydrology, Wallingford, UK. 3pp.

A replicated laboratory and field study in 2011 in South America<sup>1</sup> found that the South American weevil *Listronotus elongatus*, was the most common herbivore on floating pennywort and caused more feeding lesions on floating pennywort than on any other plant species, but field results found that the weevil did not reduce floating pennywort biomass. Other species found to feed on floating pennywort included mining flies of genus *Monochaetoscinella* and *Hydrellia*, and the larvae of the moth *Paracles quadrata*. When the weevils were allowed to invade a mixed patch containing floating pennywort in the field, the highest numbers of larvae and adult weevils were found on patches of floating pennywort with the highest biomass,

indicating that weevils perhaps move away from damaged plant sections and concentrate in the denser sections of the plant patch. Studies of adult weevil plant choice involved a three-stage process, beginning with a simple no-choice, cut-leaf feeding test on many plant species, followed by a whole-plant against cut-leaf no-choice test on selected species. Finally, South American weevils were allowed to invade a mixed patch containing floating pennywort, where after two months, damage was evaluated in 30 randomly selected 10 x 10 cm squares.

(1) Walsh, W. C. & Maestro, M. (2011) Water Pennywort. In *USDA-ARS-SABCL (South American Biological Control Laboratory). Annual Report, 2011, 47-53.*

### **1.1.2 Biological control using native herbivores**

- No evidence was captured on biological control of floating pennywort using native herbivores.

#### **Background**

Increasing the numbers of a native species, normally an arthropod, can increase the level of foraging to levels higher than normally endured by the target (Gassmann *et al.* 2006). This can potentially be used as a means of controlling invasive plants.

Gassmann A., Cock M.J.W., Shaw R. & Evans H.C. (2006) The potential for biological control of invasive alien aquatic weeds in Europe: a review. *Hydrobiologia*, 570, 217-222.

### **1.1.3 Biological control using fungal-based herbicides**

- No evidence was captured on biological control of floating pennywort using fungal-based herbicides.

#### **Background**

Application of a mass-produced, formulated product made of microorganisms can potentially be used as a means of controlling invasive plants (Gassmann *et al.* 2006). The US National Fungus Collections Database lists the fungal microorganisms found associated with floating pennywort *Hydrocotyle ranunculoides*, which include *Cercospora hydrocotyles*, *Entyloma fimbriatum*, *Entyloma hydrocotyles*, *Physoderma hydrocotylidis* and *Puccinia hydrocotyles* (Farr & Rossman 2011).

Farr, D. F. & Rossman, A.Y. (2011) Fungal Databases. Systematic Mycology and Microbiology Laboratory, ARS, USDA, USA.

Gassmann A., Cock M.J.W., Shaw R. & Evans H.C. (2006) The potential for biological control of invasive alien aquatic weeds in Europe: a review. *Hydrobiologia*, 570, 217-222.

#### 1.1.4 Physical removal

- A study in Western Australia<sup>1</sup> found that following a two-week program of physical removal of floating pennywort, the rate of growth exceeded the rate of removal.
- A study in the UK<sup>2</sup>, found that removal using a mechanical digger and monthly picking by hand greatly reduced the cover of floating pennywort but did not completely eradicate it.

#### Background

Manual or mechanical removal of floating pennywort *Hydrocotyle ranunculoides* with manual removal of any visible remaining fragments may offer a tool for localised population eradication. For example, it is reported that floating pennywort has been eradicated from the upper reaches of the River Chelmer and the River Lee by removing as much plant biomass as possible then handpicking remaining fragments (Newman & Duenas 2010). It is important to carefully manage any mechanical removal efforts as floating pennywort demonstrates high regeneration capacity, and is able to regenerate from small shoot fragments (Hussner 2008).

Hussner, A. (2008) Ökologische und ökophysiologische Charakteristika aquatischer Neophyten in NordrheinWestfalen. Dissertation, Heinrich-Heine-Universität, Düsseldorf, 192 pp.

Newman J.R. & Duenas M.A. (2010) Information sheet: control of floating pennywort (*Hydrocotyle ranunculoides*). Centre for Ecology & Hydrology, Wallingford, UK. 3pp.

A before-and-after study from 1991 to 1992 in a river in Western Australia<sup>1</sup> found that a two-week program of physical removal did not reduce floating pennywort *Hydrocotyle ranunculoides* biomass. Following the removal program in late 1991, by September 1992 the estimated biomass of floating pennywort in the river had increased from an initial 175 tonnes to 420 tonnes. Control attempts in 1991 involved a two-week programme of physical removal by cutting the floating mats of floating pennywort with sickles from small boats. The mats were then pushed by small boats to an aquatic harvester, floated to the bank and removed by a backhoe. Follow up maintenance control was continued until January 1992, when growth rates exceeded the rate of removal.

A study in 2005-2006 by the Broads Authority at Gillingham Marshes, Suffolk, UK<sup>2</sup>, found that removal of floating pennywort *Hydrocotyle ranunculoides* using a mechanical digger and extensive hand picking, along with monitoring, greatly reduced the cover of floating pennywort but did not completely eradicate it. Hand-picking was undertaken at least once a month (usually every fortnight) throughout the growing season (March – September 2005-2006 ongoing). In addition, a mesh grid was added to the upstream end of the water pump at Gillingham Marshes to try to prevent floating fragments from entering and infesting the River Waveney, adjacent to the marshes. To dispose of the pennywort, it was piled on the site and monitored for regrowth. Monthly monitoring of the pile was undertaken and if signs of growth were observed, they were sprayed with the herbicide glyphosate.

(1) Ruiz-Avila R.J. & Klemm V.V. (1996) Management of *Hydrocotyle ranunculoides* L.f., an aquatic invasive weed of urban waterways in Western Australia. *Hydrobiologia*, 340, 187-190.

(2) Kelly A. (2006) Removal of invasive floating pennywort *Hydrocotyle ranunculoides* from Gillingham Marshes, Suffolk, England. *Conservation Evidence*, 2006, 52-53.

### 1.1.5 Chemical control using herbicides

- A controlled, replicated study in the UK<sup>1</sup> found that the herbicide 2,4-D amine applied at 4.2 kg/ha achieved near to 100% mortality, compared with the herbicide glyphosate applied at 2.2 kg active ingredient/ha (without an adjuvant) which achieved negligible mortality.

#### Background

Application of chemical herbicides may offer a localised tool for management of floating pennywort *Hydrocotyle ranunculoides* provided regulatory approvals are in place. It is possible that efficacy will be enhanced by use of agents called adjuvants to bond the herbicide to the leaf surface, and the most effective adjuvants may show seasonality. For example, an information sheet published in the UK suggests that applications of 4-6 L/ha in 200 L water with an adjuvant called TopFilm at 1.2 L/ha work up to the middle of July whereas applications of 4-6 L/ha with a Codacide Oil adjuvant work from July onwards (Newman & Duenas 2010).

Newman J.R. & Duenas M.A. (2010) Information sheet: control of floating pennywort (*Hydrocotyle ranunculoides*). Centre for Ecology & Hydrology, Wallingford, UK. 3pp.

A controlled, replicated field experiment in 1997 in the Addlestone Bourne flood relief channel England, UK<sup>1</sup> found that the herbicide 2,4-D amine achieved near to 100% mortality, compared with the herbicide glyphosate which achieved negligible mortality. The chemical 2,4-D amine applied at 4.2 kg/ha achieved 76% decrease in floating pennywort *Hydrocotyle ranunculoides* biomass and almost 100% mortality over the first four weeks of treatment. In comparison, treatment with glyphosate applied at 2.2 kg/ha (without an adjuvant) resulted in a 20% decrease in biomass over the first four weeks and negligible mortality. Two trial plots in a 65m section of the channel were marked out and subdivided into six treatment blocks. Two blocks were sprayed in 2,4-D amine, two in glyphosate, and two were left untreated in control plots. Wet weight of root and shoot material/m<sup>2</sup> was taken before treatment, and each week after treatment for four weeks following herbicide application. Percentage cover measurements were made each week until nine weeks after spraying.

(1) Newman, J.R. & Dawson, F.H. (1999) Ecology, distribution and chemical control of *Hydrocotyle ranunculoides* in the U.K. *Hydrobiologia*, 415, 295-298.

### **1.1.6 Combination treatment using herbicides and physical removal**

- A before-and-after study in Western Australia<sup>1</sup> found that a combination of cutting followed by glyphosate chemical treatment, removed floating pennywort.

#### **Background**

Removing the majority of floating pennywort *Hydrocotyle ranunculoides* biomass prior to targeted herbicide application offers a tool for localised population reduction, and can be used to prevent significant deoxygenation of the water body due to decomposition of treated plant material.

A before-and-after study from 1993 to 1994 in a river in Western Australia<sup>1</sup> found that a combination of cutting followed by glyphosate chemical treatment, removed approximately 2,000 tonnes of floating pennywort *Hydrocotyle ranunculoides*. Floating pennywort mats were cut with sickles from small boats. The boats pushed mats to a conveyor harvester and mats were then floated to the bank, where they were removed. After most of the weed had been removed, the chemical glyphosate (Tradename Roundup) was applied along the banks at a rate of 360 g/ha of active ingredient in 1993 and 450 g/ha in 1994. The amount of floating pennywort removed was estimated from the number of truckloads. The area was monitored to assess re-infestation of floating pennywort and assess water quality. No further details were available.

(1) Ruiz-Avila R.J. & Klemm V.V. (1996) Management of *Hydrocotyle ranunculoides* L.f., an aquatic invasive weed of urban waterways in Western Australia. *Hydrobiologia*, 340, 187-190.

### **1.1.7 Use of hydrogen peroxide**

- A controlled, replicated pilot study in The Netherlands<sup>1</sup>, found that hydrogen peroxide sprayed on potted floating pennywort plants resulted in curling and transparency of the leaves when applied at the highest tested concentration (30%), but this was still not sufficient to kill the plant.

#### **Background**

Treatment with hydrogen peroxide offers a potential control method for floating pennywort *Hydrocotyle ranunculoides* when applied at very high concentrations.

A controlled, replicated experiment in 2010 in greenhouses at Plant Research International in Wageningen, The Netherlands<sup>1</sup>, found that hydrogen peroxide sprayed on potted floating pennywort *Hydrocotyle ranunculoides* plants resulted in curling and transparency of the leaves when applied at the highest tested concentration (30%), but this was still not sufficient to kill the plant. Leaf

transparency and curling was visible after three hours. A 10% concentration also had clear effect but killed few of the leaves, with about half of the leaf surfaces affected. Lower concentrations had little effect. Stolones and shoot tips, though all above ground, were not affected in any treatment. The roots were immersed in the mud and not affected either. The higher concentrations (10% and 30%) were reported to be hazardous to people who carry out the spraying. Test plants were newly grown cuttings 22 days old. Five treatments were chosen: 0, 0.3, 3, 10 and 30% hydrogen peroxide in water with four replications.

(1) Joost van der Burg, W. (2010) Effect of hydrogen peroxide spraying on *Hydrocotyle ranunculoides* L.f. survival. Plant Research International, Wageningen. 9pp.

### **1.1.8 Use of liquid nitrogen**

- No evidence was captured on the use of liquid nitrogen for control of floating pennywort.

#### **Background**

Liquid nitrogen may provide a tool for localised control of floating pennywort *Hydrocotyle ranunculoides* when used in combination with other control methods. However, it is reported that some water boards in the Netherlands have tried to eradicate stands of floating pennywort using liquid nitrogen, and have found this to be unsuccessful due to persistence of regenerative plant material below the water surface (Hussner *et al.* 2012). A description of the treatment and results has not been reported; therefore it has not been possible to confirm these results (Hussner *et al.* 2012).

Hussner A., Denys L., & Van Valkenburg J.L.C.H. (2012) *Hydrocotyle ranunculoides* NOBANIS – Invasive Alien Species Factsheet, 13pp.

### **1.1.9 Flame treatment**

- A controlled, replicated, pilot experiment in 2010 in The Netherlands<sup>1</sup>, found that flame treatments of 1, 2 or 3 seconds had a significantly negative and progressive impact on floating pennywort, and a 3 second repeat treatment after 11 days proved fatal.

#### **Background**

Flame treatment may provide an effective treatment for controlling floating pennywort *Hydrocotyle ranunculoides*, especially when growing on soil, and particularly when used in early spring when the plants emerge from the embankments or as an after treatment after rigorous removal has taken place.

A controlled, replicated, pilot experiment in 2010 in greenhouses in The Netherlands<sup>1</sup>, found that flame treatments of 1, 2 or 3 seconds had a significantly negative and progressive impact on floating pennywort *Hydrocotyle ranunculoides*. Repeat treatment six hours after the first had no marked effect on the eventual recovery of the plants, but a three second repeat treatment after 11 days proved fatal. The plants were collected from the wild in Laarne, Belgium in early spring 2010 and raised in containers in the greenhouse. Photographs were taken of all treatments and estimates were made of pot surface coverage by green leaves.

(1) Joost van der Burg, W. & Michielsen, J.M. (2010) Effect of flaming on *Hydrocotyle ranunculoides* L.f. survival. Plant Research International, Wageningen. 12pp.

### **1.1.10 Excavation of banks**

- No evidence was captured on the effects of excavation of banks using a sod-cutter or 'turf-cutter' to remove floating pennywort.

#### **Background**

Excavation of banks using a sod-cutter or 'turf-cutter' is a potential mechanism for removing invasive aquatic plants. An attempt to eradicate a different invasive species called New Zealand pigmyweed *Crassula helmsii* using a sod cutter to excavate the plants from the banks was reported to be partially successful (Clarke 2009).

Clarke S. (2009) A summary of three different approaches to the treatment of non-native invasive species *Crassula helmsii* at protected sites. *Abstracts and Proceedings of the 41st Robson Meeting, 17-18 February 2009, Centre for Ecology and Hydrology, Wallingford, UK, 14-17.*

### **1.1.11 Environmental control (e.g. shading, reduced flow, reduction of rooting depth, or dredging)**

- No evidence was captured on the potential for environmental control of floating pennywort using shading, increased flow, reduction of rooting depth to below 1 metre, or dredging.

#### **Background**

There are several possible methods of environmental control of floating pennywort *Hydrocotyle ranunculoides*, none of which give a complete solution.

Shade may be an effective method of control as the plant does not establish well in shaded conditions, and may be achieved by planting trees on the south side of the water body (Newman & Duenas 2010). This is unlikely to be practical to implement on larger water bodies.

Increasing flow will restrict the growth of floating pennywort in situ but may increase the spread of the plant downstream (Newman & Duenas 2010).

Increasing rooting depth to below one metre may reduce the ability of floating pennywort to root at the margins (Newman & Duenas 2010). This, however, will not often be a feasible option. Reducing the amount of suitable rooting substrate by piling or preventing access to suitable areas by netting off sections may prove effective (Newman & Duenas 2010).

All these environmental options are likely to be expensive to implement and are untested (Newman & Duenas 2010).

It is reported that dredging does not seem to be an effective method to eradicate floating pennywort (Haury *et al.* 2010). It is also reported that survivorship of floating pennywort may be prevented by a rise in salinity (Ruiz-Avila & Klemm 1996)

Haury, J., Hudin, S., Matrat, R. & Anras, L. (2010) Manuel de gestion des plantes exotiques envahissant les milieux aquatiques et les berges du bassin Loire-Bretagne. Fédération des conservatoires d'espaces naturels, Loire-Bretagne. 136 pp.

Newman J.R. & Duenas M.A. (2010) Information sheet: control of floating pennywort (*Hydrocotyle ranunculoides*). Centre for Ecology & Hydrology, Wallingford, UK. 3pp.

Ruiz-Avila R.J. & Klemm V.V. (1996) Management of *Hydrocotyle ranunculoides* L.f., an aquatic invasive weed of urban waterways in Western Australia. *Hydrobiologia*, 340, 187-190.

### **1.1.12 Public education**

- No evidence was captured on the impact of education programmes on control of floating pennywort.

#### **Background**

Once escaped in the wild, fragmentation of floating pennywort *Hydrocotyle ranunculoides* and resultant dispersal can result from a variety of management and recreational activities. Direct sales of floating pennywort are now less common in the UK due to regulatory restrictions, although labelling can be unreliable.

## 1.2 Water primrose *Ludwigia* spp.

### Background

Water primrose *Ludwigia* spp. consists of 82 species worldwide, with the greatest diversity in South America, which is considered the centre of origin for the genus and also for the family Onagraceae. The genus contains both herbaceous and woody species, as well as aquatic types (Sears *et al.* 2006).

Creeping water primrose *Ludwigia peploides* is native to South America but has spread to North America, Africa, and Europe. In the UK, creeping water primrose is only known from a few sites and it has been eradicated from some of these (Booy *et al.* undated). Uruguayan primrose-willow *Ludwigia hexapetala*, a subspecies of large-flower primrose-willow *Ludwigia grandiflora* (Dandelot 2004), is the only other non-native species of water primrose known to occur in the UK (Booy *et al.* 2013). Large-flower primrose-willow resembles and is often confused with creeping water primrose and they often occur together in the same countries. Species identification is also often complicated by environmentally-induced changes in the appearance of many aquatic *Ludwigia* species (Lakmann & Cordes 1996). Publications therefore often mention “*Ludwigia* spp.” (EPP0 2011). In this report, we include studies referencing creeping water primrose, large-flower primrose-willow, Peruvian primrose-willow *Ludwigia peruviana*, prostrate water primrose *Ludwigia prostrata*, water primrose *Ludwigia adscendens* and *Ludwigia* spp.

*Ludwigia* spp. colonise static or slow flowing waters including ditches, canals, channels, rivers and lakes. They are also found on sediment bars on river borders and in wet meadows (Zotos *et al.* 2006). Both large-flower primrose-willow and creeping water primrose are tolerant of a wide range of conditions in terms of nutrient level, types of substrate (gravel banks or sediments), pH and water quality (Matrat *et al.* 2006).

Both creeping water primrose and large-flower primrose-willow show an intense vegetative growth (Dandelot 2004). They overwinter as standing visibly dead vegetation and the main mode of spread is by fragmentation during autumn and winter (Plant Protection Service 2011). Sexual reproduction is reported to be an important additional mechanism for winter survival and spread, especially over long distances (Plant Protection Service 2011).

Water primrose is highly detrimental to the environment in Western Europe. It can form dense rafts that displace native species and reduce light levels to plants in the water column. Water quality can deteriorate beneath the rafts as a result of deoxygenation, resulting in biodiversity declines. The plant can also impede water flow, resulting in increased flood risk, and can impede navigation and angling. Water primrose also releases biochemicals that influence the water quality throughout the year and reduce the germination and survival rates of other plant species.

There is an indigenous species of water primrose in the UK which is known as marsh-purslane *Ludwigia palustris*. It is very rare and very different in appearance (Lakmann & Cordes 1996).

Booy, O., Wade, M. & White, V. (2013) Water Primrose. Information sheet, GB Non-Native Species Secretariat, Sand Hutton, UK. 2pp.

Booy, O., Wade, M. & White, V. (undated) Creeping Water-pPrimrose. Information sheet, GB Non-Native Species Secretariat, Sand Hutton, UK. 2pp.

Dandelot S. (2004) Les *Ludwigia* spp. invasives du sud de la France: Historique, biosystematique, biologie et ecologie. PhD Thesis. Universite Paul Cezanne Aix-Marseille III, France. 207pp.

EPPO (2011) Pest risk assessment for *Ludwigia grandiflora*. European and Mediterranean Plant Protection Organization, Report number 11-16827. 46pp.

Lakmann, G. & Cordes, U. (1996) *Ludwigia palustris* (L.) Elliot in Nordrhein-Westfalen und im Raum Osnabrück. *Floristische Rundbriefe*, 30, 65–79.

Matrat, R., Anras, L., Vienne, L., Hervochon, F., Pineau, C., Bastian, S., Dutartre, A., Haury, J., Lambert, E., Gilet, H., Lacroix, P. & Maman, L. (2006) Gestion des plantes exotiques envahissantes – Guide technique. Comité des Pays de la Loire de gestion des plantes exotiques envahissantes, Agence de l'Eau Loire-Bretagne, Forum des Marais atlantiques, DIREN Pays de la Loire & Conservatoire régional des rives de la Loire et de ses affluents. Second Edition, 86pp.

Plant Protection Service, Plant Research International Wageningen UR, Aquatic Ecology and Water Quality Management Group & Centre for Ecology and Hydrology Wallingford (2011) *EUPHRESKO DeCLAIM Final report A State of the art June 2011. Ludwigia grandiflora* (Michx.) Greuter & Burdet. 63pp.

Sears, A.L.W., Meisler, J. & Verdone L.N. (2006) Invasive *Ludwigia* management plan. Sonoma State University and Marin/Sonoma Mosquito and Vector Control District, Sonoma, California. 25pp.

Zotos, A., Sarika, M., Lucas, E. & Dimopoulos, P. (2006) *Ludwigia peploides* subsp. *montevidensis*, a new alien taxon for the flora of Greece and the Balkans. *Journal of Biological Research*, 5, 71-78.

## Key messages

### ***Biological control using co-evolved, host specific herbivores***

A controlled, replicated study in China, found a flea beetle caused heavy feeding destruction to the prostrate water primrose. A before-and-after study in the USA found that the introduction of flea beetles to a pond significantly reduced the abundance of large-flower primrose-willow.

### ***Biological control using native herbivores***

No evidence was found on the use of biological control of water primrose using native herbivores.

### ***Biological control using fungal-based herbicides***

No evidence was found on the use of biological control of water primrose using fungal-based herbicides.

### ***Physical removal***

A study in the USA found that hand pulling and raking water primrose failed to reduce its abundance at one site, whereas hand-pulling from the margins of a pond eradicated a smaller population of water primrose at a second site.

### ***Chemical control using herbicides***

A controlled, replicated laboratory study in the USA found that the herbicide triclopyr TEA applied at concentrations of 0.25% killed 100% of young cultivated water primrose within two months. A before-and-after field study in the UK found that the herbicide glyphosate caused 97% mortality when mixed with a non-oil based

sticking agent and 100% mortality when combined with TopFilm. A controlled, replicated, randomized study in Venezuela, found that use of the herbicide halosulfuron-methyl (Sempra) resulted in a significant reduction in water primrose coverage without apparent toxicity to rice plants.

***Combination treatment using herbicides and physical removal***

A study in the USA found that application of glyphosate and a surface active agent called Cygnet-Plus followed by removal by mechanical means killed 75% of a long-standing population of water primrose. A study in Australia found that a combination of herbicide application, physical removal, and other actions such as promotion of native plants and mulching reduced the cover of Peruvian primrose-willow by 85-90%.

***Use of hydrogen peroxide***

No evidence was found for use of hydrogen peroxide to control water primrose.

***Use of liquid nitrogen***

No evidence was found for use of liquid nitrogen to control water primrose.

***Flame treatment***

No evidence was found for use of flame treatment to control water primrose.

***Use of mats placed on the bottom of the water body***

No evidence was found for use of mats placed on the bottom of the water body to control water primrose.

***Use of a tarpaulin***

No evidence was captured on the use of tarpaulin for control of water primrose.

***Excavation of banks***

No evidence was captured on the effects of excavation of banks using a sod-cutter or 'turf-cutter' to remove water primrose.

***Environmental control (e.g. shading, altered flow, altered rooting depth, or dredging)***

No evidence was captured on the use of environmental control of water primrose using shading, altered flow, altered rooting depth, or dredging.

***Public education***

No evidence was captured on the impact of education programmes on control of water primrose.

**1.2.1 Biological control using co-evolved, host specific herbivores**

- A controlled, replicated field study in China<sup>1</sup>, found a flea beetle caused heavy feeding destruction when added to field cages containing prostrate water primrose seedlings, and was specific to the prostrate water primrose and Indian toothcup.
- A replicated, before-and-after field study in the USA<sup>2</sup> found that introduction of flea beetles to a pond significantly reduced the abundance of large-flower primrose-willow.

## Background

One-off introduction of a co-evolved, host-specific herbivore from the area of origin of the invasive pest can potentially provide sustainable control without affecting non-target indigenous plants.

Some studies have reported heavy damage caused to water primrose in the field by herbivorous insects. For example, the flea beetle *Lysathia flavipes* has been reported to cause heavy damage in the field to creeping water primrose *Ludwigia peploides*, although feeding behaviour was reportedly not specific (Cordo & DeLoach 1982a). It has also been reported that sterile grass carp *Ctenopharyngodon idella* have been used to control large-flower primrose-willow *Ludwigia grandiflora* (Manuel 1989) although grass carp are non-selective herbivores in which case they could harm native species.

Some studies reference damage caused to water primrose by certain species of insects without referencing their specificity. For example, a study published in 2000 reports that three beetles, comprising two *Lysathia* spp. and a *Macrohaltica* spp., and also the lesser vine Sphinx moth *Pholus fasciatus*, damage water primrose in Colombia but no reference is made to specificity (Cuevas Medina 2000).

In contrast, other studies appear to have found host-specific agents. For example, a specificity study in Argentina, found that adults of the weevils *Tyloderma* spp. A and *Tyloderma* spp. B appear to have creeping water primrose as their only host plant (Cordo & DeLoach 1982b).

Cordo, H. A. & DeLoach, C.J. (1982a) The flea beetle, *Lysanthia flavipes*, that attacks *Ludwigia* (water primrose) and *Myriophyllum* (parrotfeather) in Argentina. *The Coleopterists Bulletin*, 36, 298-301.

Cordo, H.A. & DeLoach, C.J. (1982b) Notes on the Weevils *Tyloderma*, *Auleutes*, and *Onychylis* That Feed on *Ludwigia* and Other Aquatic Plants in Southern South America. *The Coleopterists Bulletin*, 36, 291-297.

Cuevas Medina, A. (2000) Biological control of the weed 'palo deAgua' in rice with Coleoptera. *Arroz*, 49, 14-18.

Manuel, K.L. (1989) Proceedings of Workshop on Management of Aquatic Weeds and Mosquitoes in Impoundments March 14-15, Charlotte, North Carolina. Water Resources Research Institute, Report, 247. Water Resources Research Institute, University of North Carolina, 21-26.

A controlled, replicated field study carried out in 1985 in Jiangxi Agricultural University, China<sup>1</sup>, found the flea beetle *Altica cyanea* caused heavy feeding destruction to the prostrate water primrose *Ludwigia prostrata* and was specific to the prostrate water primrose and Indian toothcup *Rotala indica*. In field cage experiments, flea beetles decimated prostrate water primrose plants within a month. Wooden frame field cages were covered with nylon mesh on the sides and a mesh window on the top. One hundred prostrate water primrose seedlings were planted per cage. Flea beetle adults were collected from the field and zero, two, four, and eight pairs of them were released at random into each cage when young caged plants had at least nine leaves. There were three replicates of each population size on the plants. Observations were made at 10 day intervals. To test

food specificity, laboratory-based larval starvation tests were carried out on 24 different plant species at temperatures of 20-30°C.

A replicated, before-and-after field study conducted in 1994 in the USA<sup>2</sup> found that introduction of flea beetles *Lysathia ludoviciana* to a pond significantly reduced the abundance of large-flower primrose-willow *Ludwigia grandiflora*. When the beetles were introduced to the pond, the abundance of large-flower primrose-willow declined from an initial average 61 g/m<sup>2</sup> to an average of 7 g/m<sup>2</sup> from July to September. Beetles were introduced in July into a one hectare pond containing the water primrose. The mean density of flea beetles varied throughout the study from 1-12/m<sup>2</sup>. Changes in abundance of large-flower primrose-willow was monitored in six enclosures measuring 5 x 10 m.

(1)Xiao-Shui, W. (1990) *Altica cyanea* (Col: Chrysomelidae) for the biological control of *Ludwigia prostrata* (Onagraceae) in China. *Tropical Pest Management*, 36, 368-370.

(2)McGregor, M.A., Bayne, D.R., Steeger, J.G., Webber, E.C. & Reutebuch, E. (1996) The Potential for Biological Control of Water Primrose (*Ludwigia grandiflora*) by the Water Primrose Flea Beetle (*Lysathia ludoviciana*) in the Southeastern United States. *Journal of Aquatic Plant Management*, 34, 74-76.

### **1.2.2 Biological control using native herbivores**

- No evidence was found on the use of biological control of water primrose using native herbivores.

#### **Background**

Increasing the numbers of a native species can increase the level of foraging to levels higher than normally endured by the target (Gassmann *et al.* 2006). This can potentially be used as a means of controlling invasive plants. For example, a controlled, replicated field experiment on a different plant species, Parrotfeather *Myriophyllum aquaticum* in the USA found that over a two year period beaver *Castor canadensis* herbivory reduced the abundance of invasive Parrotfeather by nearly 90% (Parker *et al.* 2007).

Gassmann A., Cock M.J.W., Shaw R. & Evans H.C. (2006) The potential for biological control of invasive alien aquatic weeds in Europe: a review. *Hydrobiologia*, 570, 217-222.

Parker, J.D., Caudill, C.C. & Hay, M.E. (2007) Beaver herbivory on aquatic plants. *Oecologia*, 151, 616-625.

### **1.2.3 Biological control using fungal-based herbicides**

- No evidence was found on the use of biological control of water primrose using fungal-based herbicides.

## Background

Application of a mass-produced, formulated product made of microorganisms can potentially be used as a means of controlling invasive plants (Gassmann *et al.* 2006). For example, anecdotal evidence suggests that certain strains of a plant pathogenic fungus *Glomerella cingulata* may be effective against water primrose *Ludwigia adscendens*, St. John's wort *Hypericum perforatum* and other plant species such as Koster's Curse *Clidemia hirta* and Needlebush *Hakea sericea* (Jensen 1992).

Gassmann A., Cock M.J.W., Shaw R. & Evans H.C. (2006) The potential for biological control of invasive alien aquatic weeds in Europe: a review. *Hydrobiologia*, 570, 217-222.

Jensen K.I.N. (1991) The use of *Colletotrichum* species in biological weed control. Proceedings of the 38th annual meeting of the Canadian Pest Management Society, Fredericton, New Brunswick, Canada, 27-31 July 1991, 118-120.

### 1.2.4 Physical removal

- A study in the USA<sup>1</sup> found that hand pulling and raking water primrose failed to reduce its abundance, whereas hand-pulling from the margins of a pond eradicated a smaller population of water primrose.

## Background

Manual or mechanical removal of water primrose *Ludwigia* spp. with manual removal of any visible remaining fragments may offer a tool for localised population eradication.

However, it has been shown that water primrose demonstrates high regeneration capacity, and an ability to regenerate from small shoot fragments with a node with or without leaves (Hussner 2008). Therefore, it is important to carefully manage any mechanical removal program to prevent regenerative fragments from spreading to new locations.

Manual or mechanical removal programmes have been met with mixed success. For example, a review reports that effective management of large-flower primrose-willow *Ludwigia grandiflora* for eradication is relatively difficult, and mechanical removal tends to create viable fragments which can spread to new areas or recolonise existing managed sites (Plant Protection Service *et al.* 2011). A study found that mechanical harvesting, followed by several manual finishes over a two year period, eradicated large-flower primrose-willow. It was noted that ground realities, including inaccessible sites for heavy equipment, and difficulty observing the species, can strongly affect the feasibility (Legrand 2002). One study reported that in the early stages of water primrose *Ludwigia* spp. colonisation, removal by hand is usually possible, and that mechanised removal is only necessary when the plant has become well established (Thiébaud 2007).

- Hussner, A. (2008) Ökologische und ökophysiologische Charakteristika aquatischer Neophyten in Nordrhein-Westfalen. Ph.D. Dissertation, Heinrich-Heine-Universität Düsseldorf, 192 pp.
- Plant Protection Service, Plant Research International Wageningen UR, Aquatic Ecology and Water Quality Management Group & Centre for Ecology and Hydrology Wallingford (2011) *EUPHRESKO DeCLAIM Final report A State of the art June 2011. Ludwigia grandiflora* (Michx.) Greuter & Burdet. 63pp.
- Legrand C., 2002. Guide technique : pour contrôler la prolifération des jussies (*Ludwigia* spp.) des zones humides méditerranéennes. Agence Méditerranéenne de l'Environnement, Montpellier, France. 68pp.
- Thiébaud, G. (2007) Non-indigenous aquatic and semiaquatic plant species in France. In: Gherardi, F. (Ed.) *Biological invaders in inland waters: profiles, distribution and threats*. Springer, Dordrecht, pp. 209–229.

A study in 2005 in a managed wetland in the Laguna de Santa Rosa, California, USA<sup>1</sup>, found that hand pulling and raking water primrose *Ludwigia* sp. failed to reduce its abundance, whereas hand-pulling from the margins of a pond in the Laguna Wetland Preserve Sebastopol successfully eradicated a smaller population of water primrose. Attempts to reduce the coverage of primrose in the Laguna de Santa Rosa, California, were wholly unsuccessful and by the end of the season water primrose covered 100% of the pond. Hand pulling and raking were carried out for 2-6 person hours/week. Workers in the Laguna Wetland Preserve at Sebastopol spent approximately 150 person hours of effort hand-pulling water primrose from pond margins.

(1) Sears, A.L.W., Meisler, J. & Verdone L.N. (2006) Invasive *Ludwigia* management plan. Sonoma State University and Marin/Sonoma Mosquito and Vector Control District, Sonoma, California. 25pp.

## 1.2.5 Chemical control using herbicides

- A controlled, replicated laboratory study in the USA<sup>1</sup> found that the herbicide triclopyr TEA applied at concentrations of 0.25% killed 100% of young cultivated water primrose within two months.
- A before-and-after field study in the UK<sup>2</sup> found that the herbicide glyphosate controlled water primrose, causing 97% mortality when mixed with a non-oil based sticking agent and 100% mortality when combined with TopFilm.
- A controlled, replicated, randomized study in Venezuela<sup>3</sup>, found that use of the herbicide halosulfuron-methyl (Semptra) resulted in a significant reduction in water primrose coverage without apparent toxicity to rice plants.

### Background

Application of chemical herbicides may offer a tool for management of water primrose *Ludwigia* spp. provided regulatory approvals are in place. For example, glyphosate has been reported to have successfully eradicated water primrose *Ludwigia* spp. at three UK sites (Renals 2010).

Adjuvants are agents that can modify the effect of chemical herbicides, thereby increasing their effectiveness. It has been reported that there is a synergistic benefit of applying an adjuvant such as TopFilm alongside glyphosate. It is reported that the combined use of herbicide and adjuvant has eradicated various small stands of large-flower primrose-willow (Newman 2008). A review also references the potential for synergistic application of glyphosate and TopFilm, reporting the most effective application rates to be 2.28 kg of glyphosate and one litre/ hectare of TopFilm, and reporting June and July to be the best time for application (Plant Protection Service et al. 2011).

Renals, T. (2010) *Ludwigia* Eradication: A Rough Model for the Future. Proceedings of the 42<sup>nd</sup> Robson Meeting, CEH, Wallingford, UK. 1pp.

Newman J R (2008). Aquatic Weed Control. Proceedings of the 40th Robson Meeting. CEH, Wallingford, UK. 1pp.

Plant Protection Service, Plant Research International Wageningen UR, Aquatic Ecology and Water Quality Management Group & Centre for Ecology and Hydrology Wallingford (2011)

*EUPHRESKO DeCLAIM Final report A State of the art June 2011. Ludwigia grandiflora* (Michx.) Greuter & Burdet. 63pp.

A controlled, replicated laboratory study conducted in 2007 in the USA<sup>1</sup> found that the herbicide triclopyr TEA (triethylamine) stopped the growth of young cultivated creeping water primrose *Ludwigia peploides* in comparison to untreated plants. Plants stopped growing and were damaged at concentrations of 0.25% by volume and above. Within two months, 0.25 % triclopyr TEA killed 100 % of treated creeping water primrose. Creeping water primrose seedlings were collected from the wild and cultivated in glasshouses. The plants were grown in water-filled 40 litre tubs containing a bed of sand for at least two months. Herbicide was applied to run off at between 0.25% and 5.00% concentration by volume. Ten plants were tested at each concentration and monitored regularly for signs of herbicide damage. Ten plants were left untreated as controls.

A before-and-after field study conducted from 2006 to 2007 in the UK<sup>2</sup> found that the herbicide glyphosate combined with a non-oil soya sticking agent, reduced the abundance of creeping water primrose *Ludwigia peploides*. After 56 days, the biomass of creeping water primrose was reduced by over 97% compared with the biomass before treatment. When glyphosate was combined with TopFilm, it reportedly killed the plant. A solution of 360g/litre of glyphosate (Roundup Pro Biactive) was applied at a rate of 6 litres/ha and the non-oil soya sticking agent was applied at a rate of 450 ml/ha. Further details of the experimental treatments, such as area treated, are not provided.

A controlled, replicated, randomized study at the National Institute for Agricultural Research, Venezuela<sup>3</sup>, found that use of the herbicide halosulfuron-methyl (Sempra) resulted in a significant reduction in water primrose *Ludwigia* spp. coverage without apparent toxicity to rice plants. Treatment of 60 g active ingredient/ha produced the highest percentage control, with average reduction in water primrose coverage of 80%. The trial was conducted in a randomized block design with five treatments and four repetitions. Treatments were undertaken in experimental plots marked with fixed 0.25m<sup>2</sup> metal frames. Sempra, formulated as water dispersible granules at a concentration of 75% halosulfuron-methyl by weight,

was applied to rice crops 26 days after sowing at the following range of doses 0, 15, 30, 45 and 60 g active ingredient/ha. The herbicide was applied with a manual sprayer. Weed coverage and rice crop quality were then evaluated.

(1) Champion, P.D., James, T.K. & Carney, E.C. (2008) Evaluation of Triclopyr triethylamine for the control of wetland weeds. *New Zealand Plant Protection*, 61, 374-377.

(2) Centre for Ecology and Hydrology (2007) Development of eradication strategies for *Ludwigia* species. *Centre for Ecology and Hydrology Defra project code PH0422*. 8pp.

(3) Suárez, L., Anzalone, A., Moreno, O. (2004) Evaluation of halosulfuron-methyl herbicide for weed control in rice (*Oryza sativa* L.). *Bioagro*, 16, 173-182.

### **1.2.6 Combination treatment using herbicides and physical removal**

- A study in California, USA<sup>1</sup>, found that application of glyphosate and a surface active agent called Cygnet-Plus followed by removal by mechanical means achieved a 75% kill rate of water primrose.
- A study in Australia<sup>2</sup>, found that a combination of herbicide application, physical removal, and other actions such as promotion of native plants and mulching, reduced the coverage of Peruvian primrose-willow by 85-90%.

#### **Background**

A combination treatment of herbicide application and physical removal offers a tool for localised population reduction, and can be used to prevent significant deoxygenation of the water body due to decomposition of treated plant material.

Numerous attempts to eradicate water primrose report that a combination treatment is much more effective than chemical treatment alone. For example, a study in the USA reported that mechanical removal alone had limited effectiveness, whereas a combined glyphosate spray/mechanical removal treatment, though most expensive, gave the most lasting control of water primrose *Ludwigia* spp. (San Francisco Estuary Institute, *et al.* 2004). In support of this, it is reported that large-flower primrose-willow *Ludwigia grandiflora* was controlled in the Marais Poitevin, France by regular management actions including both mechanical and chemical methods (EPPO 2011). In addition, it is reported that after a single application of 4.3 kgs glyphosate/ha a population of water primrose *Ludwigia* spp. almost returned to its initial invasion stage after two years (Dutartre & Oyarzabal 1993; Delbart *et al.* 2013).

San Francisco Estuary Institute (2004) Field evaluations of alternative pest control operations in California waters. San Francisco Estuary Institute, Oakland, California. 107pp.

European and Mediterranean Plant Protection Organization (2011) Pest risk assessment for *Ludwigia grandiflora*. European and Mediterranean Plant Protection Organization, Report number 11-16827. 46pp.

Dutartre, A. & Oyarzabal, J. (1993) Gestion des plantes aquatiques dans les lacs et les étangs landais. *Hydroécologie Appliquée*, 5, 43-60.

A study in 2005 in the Laguna de Santa Rosa, California<sup>1</sup> found that application of glyphosate and a surface active agent called Cygnet-Plus followed by removal by mechanical means resulted in a 75% kill rate of a long-standing population of water primrose *Ludwigia* spp. and removal of 5,388 tonnes of water primrose plants. However, in some areas of incomplete kill, there was rapid regrowth. Following the eradication attempt, there was heightened turbidity. However, intensive water quality monitoring revealed very low levels of glyphosate and associated metabolites. The herbicide was applied in July 2005 from the bank, using spray hoses located on the back of specialised vehicles. It was therefore necessary to drive over water primrose located in the flooded wetland, thereby covering some with muddy water prior to spraying. Channel areas (47 hectares) were sprayed from shore. Quantitative and qualitative vegetation monitoring were carried out before and during the project.

A study in 1996-2001 in the Botany Wetlands, Australia<sup>2</sup> found that using a combination of herbicide application and physical removal, and other actions such as promotion of native plants and mulching, reduced the infestation of Peruvian primrose-willow *Ludwigia peruviana* by 85-90%. The cover of indigenous perennial plants increased. This was facilitated by 'capping' select areas of slushy mud with additional soil suitable for plant growth. Herbicide application on single-species stands was based on 1.0% 'Bi-active' glyphosate, but for mixed stands containing desirable plants 0.6% 2,4-D amine was applied. Each year, dead weed stands were mechanically cleared and burned to remove risk of regrowth. To control Peruvian primrose-willow seedling flushes, leaf and bark mulch was added to areas cleared of water primrose, and the water level of upstream ponds was managed.

(1) Sears, A.L.W., Meisler, J. & Verdone L.N. (2006) Invasive *Ludwigia* management plan. Sonoma State University and Marin/Sonoma Mosquito and Vector Control District, Sonoma, California. 25pp.

(2) Chandrasena, N., Pinto, L. & Sim, R. (2002) Reclaiming Botany Wetlands, Sydney through integrated management of *Ludwigia peruviana* and other weeds. *Proceedings of the Thirteenth Australian Weeds Conference*, 134-137.

## **1.2.7 Use of hydrogen peroxide**

- No evidence was found for use of hydrogen peroxide to control water primrose.

### **Background**

Treatment with hydrogen peroxide may offer a potential control method for water primrose *Ludwigia* spp. although studies on other species suggest that it may not be feasible. For example, a controlled, replicated pilot experiment on floating pennywort *Hydrocotyle ranunculoides* in 2010 in greenhouses in The Netherlands, found that hydrogen peroxide sprayed on potted floating pennywort plants only had a marked effect when applied at the highest tested concentration (30%), and that this was still not sufficient to kill the plant (Joost van der Burg 2010).

Joost van der Burg, W. (2010) Effect of hydrogen peroxide spraying on *Hydrocotyl ranunculoides* L.f. survival. Plant Research International, Wageningen. 9pp.

### **1.2.8 Use of liquid nitrogen**

- No evidence was found for use of liquid nitrogen to control water primrose.

#### **Background**

Liquid nitrogen may offer a potential control method for localised control of water primrose *Ludwigia* spp. when used in combination with other control methods. For certain other species however, this has been found not to be feasible. For example, a review on floating pennywort *Hydrocotyle ranunculoides* in the Netherlands found liquid nitrogen only killed parts of the floating pennywort plant above water, leaving the bulk of material underwater undamaged (Plant Protection Service et al. 2011).

Plant Protection Service, Plant Research International Wageningen UR, Aquatic Ecology and Water Quality Management Group & Centre for Ecology and Hydrology Wallingford (2011) *EUPHRESKO DeCLAIM Final report A State of the art June 2011. Ludwigia grandiflora* (Michx.) Greuter & Burdet. 63pp.

### **1.2.9 Flame treatment**

- No evidence was found for use of flame treatment to control water primrose.

#### **Background**

Flame treatment may provide an effective treatment for controlling water primrose *Ludwigia* spp. especially when growing on soil, and particularly when used as the plants emerge from the embankments or as an after treatment after a rigorous removal has taken place. It has been reported that dead water primrose stands can be removed by burning (Chandrasena *et al.* 2002). In addition, a controlled, replicated pilot experiment on a different invasive plant, floating pennywort *Hydrocotyle ranunculoides* found that flame treatments of 1, 2 or 3 seconds had a significantly negative, and progressive impact and a 3 second repeat treatment after 11 days proved fatal (Joost van der Burg & Michielsen 2010).

Chandrasena, N., Pinto, L. & Sim, R. (2002) Reclaiming Botany Wetlands, Sydney through integrated management of *Ludwigia peruviana* and other weeds. *Proceedings of the Thirteenth Australian Weeds Conference*, 134-137.

Joost van der Burg, W. & Michielsen, J.M. (2010) Effect of flaming on *Hydrocotyle ranunculoides* L.f. survival. Plant Research International, Wageningen. 12pp.

### **1.2.10 Use of mats placed on the bottom of the water body**

- No evidence was found for use of mats placed on the bottom of a water body to control water primrose.

#### **Background**

Use of mats placed on the bottom of a water body may provide a mechanism for control of small localised populations of water primrose *Ludwigia* spp. A study reported a successful attempt to eradicate a different species, swamp stonecrop *Crassula helmsii*, from small localised areas using mats comprising of black plastic sheeting (Leach & Dawson 1999).

Leach, J. & Dawson, H. (1999) *Crassula helmsii* in the British Isles – an unwelcome invader. *British Wildlife*, 10, 234-239.

### **1.2.11 Use of a tarpaulin**

- No evidence was captured on the use of tarpaulin for control of water primrose.

#### **Background**

Use of a tarpaulin may provide a mechanism for control of small localised populations of water primrose *Ludwigia* spp.

### **1.2.12 Excavation of banks**

- No evidence was captured on the effects of excavation of banks using a sod-cutter or 'turf-cutter' to remove water primrose.

#### **Background**

Excavation of banks using a sod-cutter or 'turf-cutter' is a potential mechanism for removing invasive aquatic plants. A study reported a partially successful attempt to eradicate a different species, swamp stonecrop *Crassula helmsii*, using a sod cutter to excavate the plants from the banks (Clarke 2009).

Clarke S. (2009) A summary of three different approaches to the treatment of non-native invasive species *Crassula helmsii* at protected sites. *Abstracts and Proceedings of the 41st Robson Meeting, 17-18 February 2009, Centre for Ecology and Hydrology, Wallingford, UK, 14-17.*

### **1.2.13 Environmental control (e.g shading, altered flow, altered rooting depth, or dredging)**

- No evidence was captured on the use of environmental control of water primrose using shading, altered flow, altered rooting depth, or dredging.

#### **Background**

Environmental control which includes alterations to shade, water flow speed, rooting depth, and dredging, may provide useful tools for control of water primrose *Ludwigia* spp., particularly in combination with other treatments.

Promotion of competitive, perennial, native plants can also help reclaim water primrose infested habitats. For example, during attempts from 1996-2001 to control Peruvian primrose-willow *Ludwigia peruviana* in the Botany Wetlands, Australia, it was found that once the original Peruvian primrose-willow infestations were controlled, facilitation of competitive plants, and mulching with leaf and bark mulch, drastically reduced Peruvian primrose-willow recolonization (Chandrasena *et al.* 2002).

Chandrasena, N., Pinto, L. & Sim, R. (2002) Reclaiming Botany Wetlands, Sydney through integrated management of *Ludwigia peruviana* and other weeds. *Proceedings of the Thirteenth Australian Weeds Conference*, 134-137.

### **1.2.14 Public education**

- No evidence was captured on the impact of education programmes on control of water primrose.

#### **Background**

Once escaped in the wild, fragmentation of water primrose *Ludwigia* spp. and resultant dispersal can result from a variety of management and recreational activities. Teaching users how to clean equipment in a way that decreases the chance of transmission is one way to lessen the impact of human-mediated transport. Educating the public about the dangers this plant poses outside of its native range may also help reduce the number of new introductions.

Numerous educational campaigns have been directed at informing the public about the danger of aquatic invasive species in countries in Europe to which water primrose poses a threat. Governmental organizations disseminate educational materials about the identification and control of *Ludwigia* spp.

### 1.3 Skunk cabbage *Lysichiton americanus*

#### Background

Skunk cabbage *Lysichiton americanus* is a yellow-flowered herbaceous perennial plant also known as American skunk cabbage or yellow skunk cabbage. It is native to Southern Canada and the United States. It grows in the transition zone of terrestrial, semi-aquatic and aquatic habitats like swamps, fens, wet meadows, marshy and alluvial woodlands, along streams, riverbanks, lakesides and ponds, and it has no specific site condition requirements except the presence of saturated organic soils (Scientific Panel on Plant Health 2007).

Skunk cabbage has been introduced throughout Europe; however, although it is not common in its introduced range, it may locally dominate vegetation (CABI 2015). Skunk cabbage can form dense layers of vegetation which exclude light, and thus affect biodiversity in ecologically sensitive wetland habitats (EPPO 2009). A UK survey found strong and highly significant negative relationships between skunk cabbage cover and the diversity of other species (Sanderson 2013).

Skunk cabbage grows at a slow rate but can live for up to 75 years (Rosendahl 1911). It reproduces almost exclusively by seeds, with an estimated 300-650 seeds produced per spadix (Alberternst & Nawrath 2002). With maturity of the seeds, the spadix becomes fragile, disconnects from the flower stalk and falls to the ground, with most seeds germinating directly next to the mother plant (EPPO 2009). A large seed bank can build up in the soil, remaining viable for at least eight years. However, only plants of three years or older produce seeds (EPPO 2009).

Seeds may be dispersed downstream along waterways, however it is considered that the spread by natural means is not rapid but may occur occasionally when adequate stream flows and seed maturity coincide (EPPO 2009). It is considered that spread by fragmentation of rhizomes is unlikely to occur by natural means, in addition to which the plant does not seem to move easily to new waterbodies and water catchments without human assistance (EPPO 2009).

Alberternst B. & Nawrath S. (2002) *Lysichiton americanus* Hultén & St. John neu in Kontinental-Europa. Bestehen Chancen für die Bekämpfung in der Frühphase der Einbürgerung? [*Lysichiton americanus* Hultén & St. John new in Continental Europe. Is there a chance for control in the early phase of naturalization?]. *Neobiota*, 1, 91-99.

CABI (2015) Datasheet on *Lysichiton americanus* (American skunk cabbage). CABI Invasive Species Compendium. 16 pp.

European and Mediterranean Plant Protection Organization (2009) Report of a Pest Risk Analysis for *Lysichiton americanus*. European and Mediterranean Plant Protection Organization Report Number 09-15078 rev, 61 pp.

Rosendahl C.O. (1911) Observation on the morphology of the underground stems of *Symplocarpus* and *Lysichiton*, together with some notes on geographical distribution and relationship. *Minnesota Botanical Studies*, 4, 137-152.

Scientific Panel on Plant Health (2007) Opinion of the Scientific Panel on Plant Health on the pest risk analysis made by EPPO on *Lysichiton americanus* Hultén & St. John (American or yellow skunk cabbage). *The EFSA Journal*, 539, 1-12.

## **Key messages**

### ***Biological control using co-evolved, host specific herbivores***

No evidence was captured on biological control of skunk cabbage using co-evolved, host specific herbivores.

### ***Biological control using native herbivores***

No evidence was captured on biological control of skunk cabbage using native herbivores.

### ***Biological control using fungal-based herbicides***

No evidence was captured on biological control of skunk cabbage using fungal-based herbicides.

### ***Physical removal***

Two studies in Switzerland and the Netherlands, reported effective removal of recently established skunk cabbage plants using physical removal, one reporting removal of the entire stock within five years. A third study in Germany reported that after four years of a twice yearly full removal programme, a large number of plants still needed to be removed each year.

### ***Chemical control using herbicides***

Two studies in the UK found that application of the chemical 2,4-D amine appeared to be successful in eradicating skunk cabbage stands. One of these studies also found glyphosate eradicated skunk cabbage. However, a study in the UK found that glyphosate did not eradicate skunk cabbage, but resulted in only limited reduced growth of plants.

### ***Combination treatment using herbicides and physical removal***

No evidence was found for use of combination treatment using herbicides and physical removal to control skunk cabbage.

### ***Use of hydrogen peroxide***

No evidence was found for use of hydrogen peroxide to control skunk cabbage.

### ***Use of liquid nitrogen***

No evidence was found for use of liquid nitrogen to control skunk cabbage.

### ***Use of flame treatment***

No evidence was found for use of flame treatment to control skunk cabbage.

### ***Use of a tarpaulin***

No evidence was found for use of a tarpaulin to control skunk cabbage.

### ***Environmental control (e.g. shading, or promotion of native plants)***

No evidence was captured on the potential for environmental control of skunk cabbage using shading or promotion of competitive native plants.

### ***Public education***

No evidence was captured on the impact of public education programmes on control of skunk cabbage.

### 1.3.1 Biological control using co-evolved, host specific herbivores

- No evidence was captured on biological control of skunk cabbage using co-evolved, host specific herbivores.

#### Background

One-off introduction of a co-evolved, host-specific species from the area of origin of the invasive pest can potentially provide sustainable control without affecting non-target native plants. In its native range, skunk cabbage *Lysichiton americanus* is eaten by black tailed deer *Odocoileus hemionus* (Gillingham *et al.* 1997) and by Grizzly bear *Ursus arctos* (Gyug *et al.* 2004). However, the fruits and seeds have not been reported to be eaten by animals in its introduced range (EPPO 2009).

European and Mediterranean Plant Protection Organization (2009) Report of a Pest Risk Analysis for *Lysichiton americanus*. European and Mediterranean Plant Protection Organization Report Number 09-15078 rev, 61 pp.

Gillingham M.P., Parker K.L. & Hanley T.A. (1997) Forage intake by black-tailed deer in a natural environment. *Canadian Journal of Zoology*, 75, 1118-1128.

Gyug L., Hamilton T. & Austin M. (2004) Grizzly bear *Ursus arctos*. *Accounts and Measures for Managing Identified Wildlife – Accounts V*. Ministry of Environment, British Columbia, Canada. 20 pp.

### 1.3.2 Biological control using native herbivores

- No evidence was captured on biological control of skunk cabbage using native herbivores.

#### Background

Increasing the numbers of a native herbivorous species, normally an arthropod, can increase the level of foraging to levels higher than normally endured by the target (Gassmann *et al.* 2006). This can potentially be used as a means of controlling invasive plants. For example, slugs, snails, ants, earwigs, flies and rove beetles have been observed to feed on skunk cabbage *Lysichiton americanus* populations in France (Lebreton 2007). It is possible that increasing numbers of these native herbivorous species could control skunk cabbage.

Gassmann A., Cock M.J.W., Shaw R. & Evans H.C. (2006) The potential for biological control of invasive alien aquatic weeds in Europe: a review. *Hydrobiologia*, 570, 217-222.

Lebreton A. (2007) Présence du Lysichite jaune ou Faux arum, *Lysichiton americanus* Hultén & St John (Araceae), en France. *Symbioses*, 20, 60– 64.

### 1.3.3 Biological control using fungal-based herbicides

- No evidence was captured on biological control of skunk cabbage using fungal-based herbicides.

#### Background

Application of a mass-produced, formulated product made of microorganisms can potentially be used as a means of controlling invasive plants (Gassmann *et al.* 2006).

Gassmann A., Cock M.J.W., Shaw R. & Evans H.C. (2006) The potential for biological control of invasive alien aquatic weeds in Europe: a review. *Hydrobiologia*, 570, 217-222.

### 1.3.4 Physical removal

- A study in Switzerland<sup>1</sup>, found that annual physical removal of recently established skunk cabbage plants over five years removed the entire stock.
- A study in the Netherlands<sup>2</sup> found that manual removal of mature skunk cabbage plants was effective for a small outbreak of a small-growing plant.
- A study in Germany<sup>3</sup>, reported that after the first four years of a twice yearly full removal programme of skunk cabbage, a large number of plants still needed to be removed each year.

#### Background

Due to the sensitive ecosystems where skunk cabbage *Lysichiton americanus* occurs, mechanical removal can be a useful method by which to control the population, particularly in the early years of infestation since only older plants of three years or older are capable of producing seeds (EPPO 2009). It is likely that the whole plant needs to be removed in order to be effective, as partial removal reportedly leads to vegetative reproduction and a high level of regeneration (Fuchs *et al.* 2003).

European and Mediterranean Plant Protection Organization (2009) Report of a Pest Risk Analysis for *Lysichiton americanus*. European and Mediterranean Plant Protection Organization Report Number 09-15078 rev, 61 pp.

Fuchs R., Kutzelnigg H., Feige B. & Keil P. (2003) Verwilderte Vorkommen von *Lysichiton americanus* Hultén & St. John (Araceae) in Duisburg und Mülheim an der Ruhr [Savaged occurrence of *Lysichiton americanus* Hultén & St. John (Araceae) in Duisburg and Muelheim an der Ruhr]. *Tuexenia*, 23, 373-379.

A study in 2003-2008 in Switzerland<sup>1</sup>, found that annual physical removal of recently established skunk cabbage *Lysichiton americanus* plants over five years removed the entire stock. One hundred plants were removed in 2003, compared with 20 plants in 2004, and only a few individual plants in each of 2007 and 2008. In 2007 and 2008, no more plants had germinated. From 2003 to 2006, two people

removed the plants by hand on an annual basis following which a monitoring programme was put in place to check for regrowth every second year. Total costs to 2009 were reportedly around €1000, declining from €500 in 2003, to just monitoring costs from 2008 onwards.

A study in 2005-2008 in the Netherlands<sup>2</sup>, found that manual removal of mature skunk cabbage *Lysichiton americanus* was effective for a small outbreak of a small-growing plant. In 2008, two plants of over one year old, and dozens of new seedlings were found and subsequently removed by volunteers. This followed an annual inspection and removal programme which started in December 2005. Following removal, skunk cabbage plants were dug up, and then buried deep in the ground. No further information was available.

A study in 2004-2008 in the Taunus region in Germany<sup>3</sup>, reports that manually removing mature skunk cabbage *Lysichiton americanus* was not effective as plants build up a seed bank which lasts for at least eight years. After the first four years of a twice yearly total removal programme, plants with leaf length in excess of 80cm were no longer found. However, a large number of plants still needed to be removed each year. In 2008, at least 3,773 skunk cabbage plants were removed in the Taunus region. The programme involved removal of all skunk cabbage stands twice each year. No further information was available.

(1) Buholzer S., pers. comm. (2009) In European and Mediterranean Plant Protection Organization . (2009) Report of a Pest Risk Analysis for *Lysichiton americanus*. European and Mediterranean Plant Protection Organization Report Number 09-15078 rev report.

(2) Rotteveel A.J.W. (2007) Initial eradication of *Lysichiton americanus* from the Netherlands. *Abstracts of the EWRS-Symposium 2007, Hamar, Norway*, p. 36.

(3) Fuchs R., Kutzelnigg H., Feige B. & Keil P. (2003) Verwilderte Vorkommen von *Lysichiton americanus* Hultén & St. John (Araceae) in Duisburg und Mülheim an der Ruhr [Savaged occurrence of *Lysichiton americanus* Hultén & St. John (Araceae) in Duisburg and Muelheim an der Ruhr]. *Tuexenia*, 23, 373-379.

### 1.3.5 Chemical control using herbicides

- A study in the UK<sup>1</sup> found that two herbicides, glyphosate and 2, 4-D Amine, both killed all skunk cabbage plants in test areas. However, another study in the UK<sup>2</sup> found that although using 2,4-D amine at 9 litres/ha, successfully eradicated skunk cabbage, using glyphosate was unsuccessful at eradicating skunk cabbage, with only limited reduction in growth of the plants.

#### Background

Application of chemical herbicides may offer a tool for management of skunk cabbage *Lysichiton americanus* provided regulatory approvals are in place.

A study in 2010, at Lymington Reedbeds, England, UK<sup>1</sup> found that herbicide sprays, glyphosate and 2, 4-D Amine, each killed skunk cabbage *Lysichiton*

*americanus*. Two months following treatment, most plants sprayed with glyphosate appeared to have been killed, whereas most of those sprayed with 2,4-D amine were found to have new shoots. However, six months following the treatments, a limited survey did not find any skunk cabbage plants, suggesting that both herbicide applications may have been successful. The site was divided into two sections. A larger downstream section was treated with glyphosate (Roundup Pro Biactive) at a rate of 6 litres/ha. A smaller, upstream section was treated with 2, 4-D Amine in an unspecified amount. Herbicide was applied by two people over a three day time period. Surveys were conducted for seven to eight weeks, then six months, after application.

A study in the UK<sup>2</sup> found that use of 2,4-D amine at a concentration of 9 litres/ha eradicated skunk cabbage *Lysichiton americanus*, whereas glyphosate did not eradicate skunk cabbage and caused only limited reduction of growth of the plants. The 2,4-D amine was applied in the month of May at a private garden in Sussex, and at Sheffield Park Garden National Trust property. Glyphosate was applied at a site in Scotland. No further information was available.

(1) Chatters C. (2010) New Forest non-native plants project report of measures undertaken to control American Skunk Cabbage during 2010. New Forest Plants Project, UK, 13 pp.

(2) European and Mediterranean Plant Protection Organization (2009) Report of a Pest Risk Analysis for *Lysichiton americanus*. European and Mediterranean Plant Protection Organization Report Number 09-15078 rev, 61 pp.

### **1.3.6 Combination treatment using herbicides and physical removal**

- No evidence was found for use of combination treatment using herbicides and physical removal to control skunk cabbage.

#### **Background**

A combination treatment of herbicide application and physical removal may offer a tool for localised population reduction and may avoid vegetative reproduction in cases where physical removal is incomplete.

### **1.3.7 Use of hydrogen peroxide**

- No evidence was found for use of hydrogen peroxide to control skunk cabbage.

## Background

Treatment with hydrogen peroxide may offer a potential control method for skunk cabbage *Lysichiton americanus*. This is as yet untested.

### **1.3.8 Use of liquid nitrogen**

- No evidence was found for use of liquid nitrogen to control skunk cabbage.

## Background

Liquid nitrogen may offer a potential control method for localised control of skunk cabbage *Lysichiton americanus* when used in combination with other control methods. This is as yet untested.

### **1.3.9 Use of flame treatment**

- No evidence was found for use of flame treatment to control skunk cabbage.

## Background

Flame treatment may provide a treatment for controlling skunk cabbage *Lysichiton americanus* in combination with a treatment that affects the roots.

### **1.3.10 Use of a tarpaulin**

- No evidence was found for use of a tarpaulin to control skunk cabbage.

## Background

Use of a tarpaulin may provide a treatment for controlling skunk cabbage *Lysichiton americanus*.

### **1.3.11 Environmental control (e.g. shading, or promotion of native plants)**

- No evidence was captured on the use of environmental control of skunk cabbage using shading or promotion of competitive native plants.

#### **Background**

Environmental control which includes alterations to shade, and promotion of competitive native plants, may help to control spread of skunk cabbage *Lysichiton americanus*.

### **1.3.12 Public education**

- No evidence was captured on the impact of public education programmes on control of skunk cabbage.

#### **Background**

Most occurrences of skunk cabbage *Lysichiton americanus* have resulted from human introduction as an ornamental plant. In addition, where skunk cabbage has been purposefully planted for example along watercourses, and along the margins of ponds and artificial lakes, management activity such as movement of soil and pond cleaning may spread the seeds, and thereby increase the spread of this non-native plant. Public education programmes can potentially be used to reduce the horticultural trade in skunk cabbage and reduce the spread of mature plants that have been introduced.

## 1.4 New Zealand pigmyweed *Crassula helmsii*

### Background

New Zealand pigmyweed *Crassula helmsii*, also known as Australian swamp stonecrop, is a succulent, perennial plant native to Australia and New Zealand. It is an invasive aquatic weed in a number of European countries and was first recorded in the wild in the UK in the 1950s. It is found in and around ponds, lakes, reservoirs, canals, and ditches, and tolerates a wide range of conditions (EPPO 2007, Lansdown 2015).

*C. helmsii* grows on the margins of water bodies, and will form extensive colonies on damp mud, such as the margins of ponds and edges of reservoirs. *C. helmsii* will also grow out into deeper water as a floating mat growing from 0.5 m above water to 3 m below water, and its vegetation can be extremely dense (Lansdown 2015). This can lead to *C. helmsii* clogging waterways, impeding navigation, and obstructing water flow and drainage leading to flooding.

The dense vegetation can also potentially shade out or smother other plants, leading to changes or reductions in the diversity and abundance of native plant species present at a site (Dawson & Warman 1987, Smith & Buckley 2015). Floating mats of *C. helmsii* can lead to deoxygenation of the water below, with possible negative impacts on native invertebrates, fish and amphibians. There is some evidence of negative effects of *C. helmsii* on reproduction in newts (Langdon *et al.* 2004, EPPO 2007), although in general evidence for the impact of *C. helmsii* on native species is limited and variable (Dawson & Warman 1987, Langdon *et al.* 2004, Ewald 2014).

Vegetative reproduction, in which stem fragments detach from parent mats to spread downstream and colonize further sites, is the most common method of dispersal. The regeneration capacity of *C. helmsii* is high, since a new plant can form from a single node fragment (Hussner 2009). Recent evidence has shown that *C. helmsii* can also reproduce via seed outside its native range (D'hondt *et al.* 2016). Like other species in its genus, *C. helmsii* has the ability to use Crassulacean acid metabolism to take up carbon dioxide, particularly in low light levels. This may increase its competitive ability in terms of growth, via increased rates of carbon fixation (Klavsen & Maberly 2009).

*C. helmsii* can be very difficult to eliminate from a site once it is well-established, and a continuing control programme lasting several years is likely to be necessary once it has become abundant at a site.

Dawson F.H. & Warman E.A. (1987) *Crassula helmsii* (T. Kirk) cockayne: Is it an aggressive alien aquatic plant in Britain? *Biological Conservation*, 42, 247-272.

D'hondt B., Denys L., Jambon W., De Wilde R., Adriaens T., Packet J. & van Valkenburg J. (2016). Reproduction of *Crassula helmsii* by seed in western Europe. *Aquatic Invasions*, 11, 125-130.

EPPO (2007) *Crassula helmsii*. *EPPO Bulletin*, 37, 225-229.

Ewald N.C. (2014) *Crassula helmsii* in the New Forest. *Final report on the status, spread and impact of this non-native invasive plant, and the efficacy of control techniques following a 3 year*

trial. Prepared on behalf of the New Forest Non-Native Plants Project, Freshwater Habitats Trust, Oxford, UK.

Hussner A. (2009), Growth and photosynthesis of four invasive aquatic plant species in Europe. *Weed Research*, 49, 506–515.

Klavsén S.K. & Maberly S.C. (2009) Crassulacean acid metabolism contributes significantly to the in situ carbon budget in a population of the invasive aquatic macrophyte *Crassula helmsii*. *Freshwater Biology*, 54, 105–118.

Langdon S.J., Marrs R.H., Hosie C.A., McAllister H.A., Norris K.M. & Potter J.A. (2004) *Crassula helmsii* in U.K. Ponds: Effects on Plant Biodiversity and Implications for Newt Conservation. *Weed Technology*, 18, 1349-1352.

Lansdown R.V. (2015) New Zealand pigmyweed *Crassula helmsii*. NNSF Factsheet.

<http://www.nonnativespecies.org/factsheet/factsheet.cfm?speciesId=1017>

Smith T. & Buckley P. (2015) The growth of the non-native *Crassula helmsii* (Crassulaceae) increases the rarity scores of aquatic macrophyte assemblages in south-eastern England. *New Journal of Botany*, 5, 192-199.

## Key messages

### **Biological control using fungal-based herbicides**

We captured no evidence on the biological control of *Crassula helmsii* using fungal-based herbicides.

### **Biological control using herbivores**

We captured no evidence on the biological control of *Crassula helmsii* using herbivores.

### **Physical control using manual/mechanical control or dredging**

We captured no evidence on the physical control of *Crassula helmsii* using mechanical or manual control or dredging.

### **Chemical control using herbicides**

Seven studies in the UK, including one replicated, controlled study, found that applying glyphosate reduced *Crassula helmsii*. Three out of four studies in the UK, including one controlled study, found that applying diquat or diquat alginate reduced or eradicated *C. helmsii*. One small trial found no effect of diquat on *C. helmsii* cover. One replicated, controlled study in the UK found dichlobenil reduced biomass of submerged *C. helmsii* but one small before-and-after study found no effect of dichlobenil on *C. helmsii*. A replicated, controlled study found that treatment with terbutryne partially reduced biomass of submerged *C. helmsii* and that asulam, 2,4-D amine and dalapon reduced emergent *C. helmsii*.

### **Use of hydrogen peroxide**

One controlled tank trial in the UK found that hydrogen peroxide did not control *Crassula helmsii*.

### **Use of liquid nitrogen**

We captured no evidence on the use of liquid nitrogen for control of *Crassula helmsii*.

### **Use hot foam**

One replicated, controlled study in the UK found that treatment with hot foam, along with other treatments, did not control *Crassula helmsii*. A before-and-after study in the UK found that treatment with hot foam partially destroyed *C. helmsii*.

***Use salt water***

Two replicated, controlled container trials and two before-and-after field trials in the UK found that seawater eradicated *Crassula helmsii*.

***Use hot water***

We captured no evidence on the use of hot water for control of *Crassula helmsii*.

***Use flame-throwers***

We captured no evidence on the use of flame-throwers for control of *Crassula helmsii*.

***Use dyes to reduce light levels***

One replicated, controlled study in the UK found that applying aquatic dye, along with other treatments, did not reduce cover of *Crassula helmsii*.

***Use lightproof barriers***

Five before-and-after studies in the UK found that covering with black sheeting or carpet eradicated or severely reduced cover of *Crassula helmsii*.

***Alter environmental conditions to control plants (e.g. shading by succession, increasing turbidity, re-profiling or dredging)***

We captured no evidence on the environmental control of *Crassula helmsii* using shading, increasing turbidity, re-shaping or re-profiling banks of waterbodies or dredging.

***Plant other species to suppress growth of Crassula helmsii***

We captured no evidence on the use of other plants to suppress growth of *Crassula helmsii*

***Use grazing***

One of two replicated, controlled studies in the UK found that excluding grazing reduce abundance and coverage of *Crassula helmsii*. The other study found that ungrazed areas had higher coverage of *C. helmsii* than grazed plots.

***Dry out waterbodies***

We captured no evidence on the effects of draining waterbodies on *Crassula helmsii*

***Bury plants***

We captured no evidence on the use of burying with soil alone to control *Crassula helmsii*

***Surround with wire mesh***

We captured no evidence on the use wire mesh to control spread of *Crassula helmsii*

***Decontamination to prevent further spread***

One controlled, replicated container trial in the UK found that submerging *Crassula helmsii* fragments in hot water led to higher mortality than drying out plants or a control.

***Public education***

We captured no evidence on the impact of education programmes on control of *Crassula helmsii*.

***Use a combination of control methods***

One before-and-after study in the UK found that covering *Crassula helmsii* with carpet, followed by treatment with glyphosate, killed 80% of the plant.

### 1.4.1 Biological control using fungal-based herbicides

- We found no evidence for the effects of biological control of *Crassula helmsii* using fungal-based herbicides.

#### Background

Application of a mass-produced product, formulated from fungal microorganisms which cause disease in the host plant, can be used as a means of controlling invasive plants (Gassmann *et al.* 2006).

A stem- and leaf-infecting plant fungal pathogen (*Colletotrichum* species) from Australia has also been identified as a possible biological control agent of *C. helmsii*, with trials achieving infection of UK biotypes (CABI 2014). Several other fungal species could potentially be used for biological control, depending on their host-specificity and effectiveness in controlling *C. helmsii* (Shaw 2013).

CABI (2014) *CABI Annual Report. Europe UK 2012*. CABI, Egham, UK.

Gassmann A., Cock M.J.W., Shaw R. & Evans H.C. (2006) The potential for biological control of invasive alien aquatic weeds in Europe: a review. *Hydrobiologia*, 570, 217-222.

Shaw, R.H. 2013. *Progress with weed biocontrol projects, CABI in the UK*. June 2013. CABI, UK.

### 1.4.2 Biological control using herbivores

- We found no evidence for the effects of biological control using specific, non-selective or native herbivores on *Crassula helmsii*.

#### Background

The introduction of a co-evolved herbivore from the area of origin of the invasive plant, which eats only the target plant species, could potentially provide sustainable control without affecting other native plants. However, *C. helmsii* has few reported natural enemies, and a biological control programme should consider consequences for closely-related, protected, native species, such as *Crassula aquatica* (Sheppard *et al.* 2006).

Some species of host-specific insects, such as chrysomelid and curculionid beetles, have been used as biological control agents of invasive aquatic weeds in several areas globally with some success (Gassman *et al.* 2006; see also 'Water primrose *Ludwigia spp.* – Biological control using co-evolved, host-specific herbivores'). Mite species which parasitize plants (eriophyid mites) have also been used for biological control of invasive plants (Smith *et al.* 2010). These mites have high host specificity and negatively affect the host plants' reproductive success, so could potentially limit the rate of spread of *C. helmsii*. The stem-mining fly *Hydrellia perplexa* has also been

investigated as a potential biological control agent, although initial results suggest impacts on *C. helmsii* may be small (Shaw 2013, CABI 2014).

Grass carp *Ctenopharyngodon idella* is another herbivorous species used to control invasive freshwater plant species (see 'Water primrose *Ludwigia spp.* – Biological control using co-evolved, host specific herbivores'). However, these fish are non-selective, and *C. helmsii* appears not to be a preferred food for this species (Dawson & Warman 1987).

CABI (2014) *CABI Annual Report. Europe UK 2012*. CABI, Egham, UK.

Dawson F.H. & Warman E.A. (1987) *Crassula helmsii* (T. Kirk) Cockayne: Is it an aggressive alien aquatic plant in Britain? *Biological Conservation*, 42, 247-272.

Gassmann A., Cock M.J.W., Shaw R. & Evans H.C. (2006) The potential for biological control of invasive alien aquatic weeds in Europe: a review. *Hydrobiologia* 570, 217-222.

Shaw, R.H. 2013. *Progress with weed biocontrol projects, CABI in the UK*. June 2013. CABI, UK.

Sheppard A.W., Shaw R.H. & Sforza R. (2006) Top 20 environmental weeds for classical biological control in Europe: a review of opportunities, regulations and other barriers to adoption. *Weed Research*, 46, 93-117.

Smith, L., de Lillo E. & Amrine Jr. J.W. (2010) Effectiveness of eriophyid mites for biological control of weedy plants and challenges for future research. *Experimental and Applied Acarology*, 51, 115-149.

### **1.4.3 Physical control using manual/mechanical control or dredging**

- We found no evidence for the effects of physical control, using manual or mechanical control or dredging, on *Crassula helmsii*.

#### **Background**

Mechanical control could be used to remove *Crassula helmsii* from affected waterways, either by removing vegetation using mechanical or manual methods, or by dredging entire waterbodies. This approach may reduce the dominance of the plant, but is often not fully effective. This is because of the difficulty of removing all the vegetation without small sections breaking off, in combination with *C. helmsii*'s ability to regenerate from any small fragments that remain (Dawson & Warman 1987). It may require drainage of a pond and removal of the top layer of soil to completely eradicate *C. helmsii* (Leach & Dawson 2000). The intervention 'Surround with wire mesh' discusses using mesh to surround treatment sites and minimize spread of disturbed plant fragments. Dredging and re-profiling waterbodies are included in the intervention 'Alter environmental conditions to control plants', and other physical approaches to removal of *C. helmsii* are discussed in 'Drying-out waterbodies' and 'Burying plants'.

Dawson F. H. & Warman E.A. (1987) *Crassula helmsii* (T. Kirk) Cockayne: Is it an aggressive alien aquatic plant in Britain? *Biological Conservation*, 42, 247-272.

Leach J. & Dawson H. (2000) Is resistance futile? The battle against *Crassula helmsii*. *Journal of Practical Ecology and Conservation*, 4, 7-17.

#### 1.4.4 Chemical control using herbicides

- Seven studies (including one replicated and controlled study) in the UK, found that applying glyphosate reduced *Crassula helmsii*<sup>1,2,3,4,5,6,7</sup>. In one before-and-after study at single site glyphosate applied in combination with diquat reduced *C. helmsii* by 98%<sup>4</sup>. Another before-and-after study at a single site found that covering *C. helmsii* with carpet before treating with glyphosate resulted in an 80% reduction in the plant<sup>7</sup>.
- Three out of four studies (including one controlled study) in the UK<sup>2,3,4,5</sup> found that applying diquat or diquat alginate reduced cover<sup>2,4,5</sup> or eradicated<sup>2</sup> submerged *C. helmsii*. One before-and-after study at a single site found that applying both diquat and glyphosate reduced *C. helmsii* by 98%<sup>4</sup>. One small, before-and-after trial<sup>3</sup> found no effect of diquat or diquat alginate on cover of *C. helmsii*.
- One out of two studies (including one replicated, controlled study) in the UK<sup>2</sup>, found that treating submerged *C. helmsii* with dichlobenil in container trials led to partial reduction in its biomass<sup>2,3</sup>. One small before-and-after field study<sup>3</sup> found no effect of dichlobenil on *C. helmsii*.
- One replicated, controlled container trial in the UK<sup>2</sup> found that treatment with terbutryne partially reduced biomass of submerged *C. helmsii*. The same study found reductions in emergent *C. helmsii* following treatment with asulam, 2,4-D amine and dalapon.

#### Background

Application of chemical herbicides may offer a tool for localized management of *C. helmsii*, provided regulatory approvals are in place. Many herbicides apart from glyphosate are no longer approved for aquatic use in Europe. Vegetation above the water may require a different approach to that below the water, meaning that more than one herbicide may be required to eradicate all *C. helmsii* from a site. The most commonly-used herbicide on emergent vegetation is glyphosate and on submerged vegetation is diquat. Additives (adjuvants) can be used to improve the efficacy of application of chemical treatments. Retreatment after initial application of herbicide is often required.

The intervention 'Surround with wire mesh' discusses the use of mesh to surround treatment sites and minimize spread of disturbed plant fragments during herbicide application.

A replicated, controlled study in 2011-2014 at waterbodies in the New Forest, UK (1) reported that treatment with herbicide reduced cover of *C. helmsii* although this was not statistically tested. Average coverage of *C. helmsii* fell from 41% before to 9% after herbicide treatment, and it was eliminated from two out of five sites. Coverage of *C. helmsii* at control sites increased from 63% to 70%. The study also found that coverage of native plant species fell from 33% to 20% at treatment sites and from 17% to 14% at control sites during the trial. The glyphosate-based

herbicide Roundup was used at five ponds at rate of 0.3 l/ha, once in autumn 2011 and twice during autumn 2013. No treatment occurred in 2012, and some ponds were only partially treated in 2013, because of high rainfall. The authors also reported that some *C. helmsii* patches were missed from treatment. *C. helmsii* coverage was assessed in five random 0.25 m<sup>2</sup> quadrats within each treatment area in winter and summer 2011-2014, and also in seven control ponds.

A controlled container experiment in 1988-1994 in southern England, UK (2) reported that control of *C. helmsii* varied between herbicide types, although no statistical tests were carried out. Treatment of low-biomass, underwater *C. helmsii* with diquat reduced biomass by 100%. Submerged *C. helmsii* biomass was also reduced by diquat-alginate (97% reduction), dichlobenil (66%) and terbutryne (48%). Glyphosate caused the greatest biomass reduction in trials on plants above the water (82%). Emergent *C. helmsii* biomass was also reduced by asulam (66%), 2,4-D amine (55%) and dalapon (51%). Trials of higher herbicide concentrations on higher biomass of *C. helmsii* showed that diquat and diquat alginate reduced biomass by at least 85% at all concentrations. However even at the highest concentration, *C. helmsii* was not eliminated. The biomass reduction caused by glyphosate was lower and more variable (between 5% and 80%). Fourteen 0.25 x 0.25 m turfs were grown in either deep or shallow water in 300 l tanks. Low-biomass trials used 13-16 kg/m<sup>2</sup> fresh weight *C. helmsii* and took place in October 1988 and summer 1989 and 1990. High-biomass trials took place in spring 1993 and 1994 using up to 50 kg/m<sup>2</sup> *C. helmsii*. Glyphosate and diquat were applied between 1x and 50x usual concentration. Trials lasted 5-12 weeks.

A small, controlled, before-and-after trial in 1992 in two lakes in West Sussex, UK (3) reported that spraying *C. helmsii* with glyphosate reduced plant height and cover, but dichlobenil and diquat had no effect (although no statistical tests were carried out). *C. helmsii* cover decreased from 100% to 60% 11 days after spraying emergent plants with glyphosate, but did not change in the control plot. Thirty five days after spraying, treated plants were shorter (4 cm) than control plants (15 cm). At the same lake, a small plot of submerged *C. helmsii* was treated with dichlobenil but this had no effect (no data provided). At the second lake, diquat alginate did not affect cover of submerged *C. helmsii* (before: 100%; 35 days after: 95%). In a second trial at the same site, diquat did not affect cover of submerged (before: 95%; 16 days after: 95%), floating (before: 70%; after: 70%) or emergent plants (before: 15-20%; after: 15%). Glyphosate with an active ingredient concentration of 2.5 kg/ha was sprayed onto leaves at three 1 x 6 m plots in August. One control plot was left untreated. Dichlobenil granules were applied at 150 kg/ha at a 1 x 4 m plot in July. Diquat alginate, containing 100 g/l active ingredient, was applied at 10 l/ha to the water surface in August. Diquat, containing 200 g/l active ingredient, was applied at 25 l/ha to shallow areas and 50 l/ha to deep areas in September. Sites were monitored twice before (June-July) and three times after (August-October) treatments.

A before-and-after trial at a single waterbody in a nature reserve in 2000-2002 in Hampshire, UK (4) reported that treatment with diquat and glyphosate herbicide nearly eradicated *C. helmsii*, although no statistical tests were carried out. The treatment cleared 98% of *C. helmsii* from the lake, and 'no major regrowth' was reported two years after treatment. The diquat-based herbicide Reglone was applied

at 50 l/ha in March 2000, and again at 35 l/ha one month later. The glyphosate-based herbicide Roundup was sprayed onto vegetation around the edge of the lake in November.

A before-and-after study in 2001-2004 at waterbodies in a nature reserve in Kent, UK (5) reported that application of either diquat-based herbicide or glyphosate killed most *C. helmsii* plants, although no statistical tests were carried out. Spraying once, using diquat on field ditches and glyphosate on the margin of a gravel pit, killed 70% of *C. helmsii*. A second application of diquat the following year was recorded as being 'partially successful' (no data given). However the authors reported that re-growth of *C. helmsii* was 'noticed annually' at sites treated with both chemicals. The diquat-based chemical Reglone was sprayed onto 50 m<sup>2</sup> ditch at a rate of 10 l for 300 m in August-September 2001 and 2002. Glyphosate was applied at an unknown concentration to a single site in August-September 2004.

A before-and-after field trial in 2004 at a single waterbody in a nature reserve in South Yorkshire, UK (6) reported that treating *C. helmsii* with glyphosate-based herbicide partially destroyed the plants, although no statistical tests were carried out. Spraying with glyphosate killed approximately 50% of *C. helmsii*, but did not eradicate it. Glyphos biactive was sprayed on exposed plants in a shallow pond at 5 l/ha in July-August 2004. No details of the size of area treated or monitoring were provided.

A before-and-after study in 2002-2003 at a single pond in Surrey, UK (7) reported that covering plants with carpet strips and then applying glyphosate reduced the area of *C. helmsii*, although no statistical tests were carried out. Approximately 80% of *C. helmsii* was killed, although it is not clear whether this was a result of herbicide application or was due to the use of carpet to prevent light reaching the plant. In autumn 2002, strips of carpet were placed over *C. helmsii* wherever possible on the pond edge and in shallow water. In July-August 2003 the carpet was removed and the glyphosate-based herbicide Roundup was applied twice (concentration not given). *C. helmsii* cover was assessed in June 2004.

(1) Ewald N.C. (2014) *Crassula helmsii* in the New Forest. Final report on the status, spread and impact of this non-native invasive plant, and the efficacy of control techniques following a 3 year trial. Prepared on behalf of the New Forest Non-Native Plants Project, Freshwater Habitats Trust, Oxford, UK.

(2) Dawson F. H. (1996) *Crassula helmsii*: Attempts at elimination using herbicides. *Hydrobiologia*, 340, 241-245.

(3) Child, L.E. & Spencer-Jones D. (1995) *Treatment of Crassula helmsii – a case study*. Pp 195-202 in Pysek P., Prach K., Rejmanek M. & Wade M. (eds) *Plant Invasions: General Aspects and Special Problems*. International Workshop on Plant Invasions. Sep 16-19, 1993, Kostelec nad cernymi lesy, Czech Republic.

(4) Stone I. (2002) War against *Crassula* – one year on. *Enact*, 9-10.

(5) Gomes B. (2005) Controlling New Zealand pygmyweed *Crassula helmsii* in field ditches and a gravel pit by herbicide spraying at Dungeness RSPB Reserve, Kent, England. *Conservation Evidence*, 2, 6

(6) Bridge T. (2005) Controlling New Zealand pygmyweed *Crassula helmsii* using hot foam, herbicide and by burying at Old Moor RSPB Reserve, South Yorkshire, England. *Conservation Evidence*, 2, 33-34.

(7) Anonymous (2004) Chemical control of Australian swamp stonecrop (New Zealand Pygmy Weed) *Crassula helmsii*. *The National Trust Conservation Newsletter*, 8, 2-3.

### 1.4.5 Use hydrogen peroxide

- One controlled study in the UK<sup>1</sup> using tank trials found that hydrogen peroxide did not control *Crassula helmsii*.

#### Background

Hydrogen peroxide is a chemical that has herbicidal properties, but does not produce harmful chemicals that persist in the environment (Fowler & Barrett 1986). Therefore it could be used to locally control *C. helmsii*, with the advantage that it is non-toxic and leaves no residue.

Fowler M.C. & Barrett P.R.F. (1986) *Preliminary studies on the potential of hydrogen peroxide as an algicide on filamentous species*. Proceedings EWRS/AAB 7th International Symposium on Aquatic Weeds, 113-118.

A controlled tank trial in 1988-1989 in southern England, UK (1) reported that hydrogen peroxide did not reduce the biomass of *C. helmsii*, although no statistical tests were carried out. Tank trials using low concentrations of hydrogen peroxide did not reduce the mass of emergent or submerged *C. helmsii* (data not given). Treatment with a higher concentration of hydrogen peroxide led to a 24% reduction in emergent vegetation, but a 45% increase in submerged vegetation compared to a control. *C. helmsii* was grown either in deep water as submerged plants, or in shallow water as emergent plants, in 300 l tanks. In low concentration trials hydrogen peroxide was applied to submerged and emergent vegetation at 20 g/m<sup>2</sup> or 100 g/m<sup>2</sup> in autumn 1988. High concentration trials used 250 g/m<sup>2</sup> in spring 1989. Final biomass was measured after ten weeks.

(1) Dawson, F. H. & Henville P. (1991) *An investigation of the control of Crassula helmsii by herbicidal chemicals (with interim guidelines on control)*. Report to the Nature Conservancy Council (GB).

### 1.4.6 Use of liquid nitrogen to kill plants

- We found no evidence for the effects of treating *Crassula helmsii* with liquid nitrogen.

#### Background

Liquid nitrogen (-196 °C) can kill vegetation by freezing, and has the advantage that no harmful residues are left in the environment. It could therefore be used for targeted treatment of localized patches of *C. helmsii*. However, a small trial suggested that only parts of the plant that came into direct contact with liquid nitrogen were killed (Leach & Dawson 2000, no data given). Dense mats of *C. helmsii* are therefore likely to insulate all but the top layer of vegetation from the effects of freezing. Specialist training is also required for operators applying liquid nitrogen.

Leach J. & Dawson H. (2000) Is resistance futile? The battle against *Crassula helmsii*. *Journal of Practical Ecology and Conservation*, 4, 7-17.

### 1.4.7 Use hot foam

- One replicated, controlled study in the UK<sup>1</sup> found that treatment with hot foam, along with other treatments, did not reduce cover of *Crassula helmsii*. A before-and-after study in the UK<sup>2</sup> found that applying hot foam partially destroyed *C. helmsii*.

#### Background

Hot foam has been used proposed as an approach for controlling vegetation. A biodegradable, plant-based foam allows heat to remain in contact with plant surfaces for a longer period than using hot water, with the aim of rupturing plant cells and killing the plant.

The use of hot water to control *C. helmsii* is discussed in 'Use hot water'.

A replicated, controlled study in 2011-2014 at waterbodies in the New Forest, UK (1) reported that treatment with hot foam did not reduce cover of *C. helmsii*, although this was not tested statistically. Average coverage of *C. helmsii* was 56% before and 60% at the end of the hot foam treatment, compared to 63% and 70% respectively at control sites. The study also found that coverage of native plant species fell from 31% to 14% at treatment sites and from 17% to 14% at control sites over the trial period. A biodegradable agent composed of plant oils and sugars was applied as a very hot foam (above 97 °C for 2 s) to five ponds twice during autumn 2011 and autumn 2013. No treatment occurred in 2012, and two ponds were only partially treated in 2011 and 2013, because of high rainfall. Aquatic dye treatment was additionally applied to these two ponds. *C. helmsii* coverage was assessed in five random 0.25 m<sup>2</sup> quadrats within each treatment area in winter and summer from 2011-2014, and also in seven control ponds.

A before-and-after study in 2003 at waterbodies in a nature reserve in South Yorkshire, UK (2) reported that spraying with hot foam partially destroyed *C. helmsii*, although statistical tests were not carried out. Approximately 50% of *C. helmsii* was killed by the treatment, but only the top layers of the plant were affected. Biodegradable 'Waipuna' hot foam, an organic compound of corn and coconut sugars, was sprayed three times between September and November 2003. No information about the number or size of waterbodies treated, or monitoring was provided.

(1) Ewald N.C. (2014) *Crassula helmsii* in the New Forest. *Final report on the status, spread and impact of this non-native invasive plant, and the efficacy of control techniques following a 3 year trial*. Prepared on behalf of the New Forest Non-Native Plants Project, Freshwater Habitats Trust, Oxford, UK.

(2) Bridge T. (2005) Controlling New Zealand pygmyweed *Crassula helmsii* using hot foam, herbicide and by burying at Old Moor RSPB Reserve, South Yorkshire, England. *Conservation Evidence*, 2, 33-34.

### 1.4.8 Use salt water

- Two replicated, controlled container trials<sup>1,2</sup> and two before-and-after field trials<sup>1,3</sup> in the UK found that seawater eradicated *Crassula helmsii*.

#### Background

The tolerance of freshwater plant species, such as *C. helmsii*, to salt water is variable, but at high salinities it is likely to have adverse effects. However, these effects are likely to extend to native co-occurring plants.

A replicated, controlled container trial and a small, replicated, before-and-after field trial in 2006–2009 at a grazing marsh in Essex, UK (1) reported that flooding with seawater eradicated *C. helmsii*, although no statistical tests were carried out. In the container trial, *C. helmsii* turfs grown in seawater were described as ‘appeared to be dead’ after five months, but those in freshwater and brackish water were still growing. In the field trial no *C. helmsii* was observed at the end of the seawater flooding period nor 9-11 months after seawater was drained. In the container trials, 15 x 15 cm *C. helmsii* turfs were placed in 15 l containers and checked every 6-7 days. Four were covered with brackish water (2,000 µS electrical conductance), four with seawater (30,000 µS) and four with freshwater. Field trials were carried out at two sites reported as having ‘abundant and widespread’ *C. helmsii*. An 8 ha field and a 120 ha field were flooded with seawater to 5 cm above the usual winter water level from April 2006-January 2007 and January-December 2009 respectively. *C. helmsii* was surveyed between August 2007 and August 2010.

A replicated, controlled study in containers in 2011 in Dorset, UK (2) found that increased water salinity led to decreased growth rate in *C. helmsii*. *C. helmsii* growth rates were highest in the freshwater control (0.05 g/day), and declined as the salinity of the water increased (0.015 g/day in 2 parts per thousand, 0.005 g/day in 4 ppt and a loss of 0.010 g/day in 8 ppt). After 31 days *C. helmsii* in the 8 ppt salinity treatment had died. Nutrient concentration did not affect growth rate. Ten gram samples of *C. helmsii* were grown outdoors in 5 l plastic containers in September-October and sampled after 31 days. Three salinities and a freshwater control were tested at three different nutrient concentrations, and each treatment combination was replicated four times.

A before-and-after trial in a coastal grazing marsh in Hampshire, UK (3) reported that inundation with salt water nearly eradicated *C. helmsii*, although no statistical tests were carried out. One year after the seawater treatment, 99% of *C. helmsii* had been killed. The plant only remained in places not reached by the seawater. Two

years after the treatment the salt level had reduced to 2 ppt, but the original flora had not returned. The site was flooded with seawater in July 2008 and then naturally filled with rainwater in autumn 2008. *C. helmsii* cover was assessed in 2009. No details about the depth of flooding or size of site were provided.

(1) Charlton P.E., Gurney M. & Lyons G. (2010) Large-scale eradication of New Zealand pygmy weed *Crassula helmsii* from grazing marsh by inundation with seawater, Old Hall Marshes RSPB reserve, Essex, England. *Conservation Evidence*, 7, 130-133.

(2) Dean C., Day J., Gozlan R.E., Green I., Yates B. & Diaz A. (2013) Estimating the minimum salinity levels for the control of New Zealand pygmyweed *Crassula helmsii* in brackish water habitats. *Conservation Evidence*, 10, 89-92.

(3) EPPO (2014) *PM 9/19 (1) Invasive alien aquatic plants*. EPPO Bulletin, 44, 457-471.

### 1.4.9 Use hot water

- We found no evidence on the use of hot water to control *Crassula helmsii*.

#### Background

Treatment with hot water can kill *Crassula helmsii* (Anderson *et al.* 2015), and could potentially be used to control small local patches of the plant. However, if it opens up bare patches of earth, this may provide suitable conditions for recolonization by *C. helmsii* (Ewald 2014).

Treatment of equipment with hot water to prevent spread is discussed in 'Decontamination to prevent spread'. Studies that used hot foam to control *C. helmsii* are described in 'Use hot foam'.

Anderson L.G., Dunn A.M., Rosewarne P.J. & Stebbing P.D. (2015) Invaders in hot water: a simple decontamination method to prevent the accidental spread of aquatic invasive non-native species. *Biological Invasions*, 17, 2287-2297.

Ewald N.C. (2014) *Crassula helmsii* in the New Forest. Final report on the status, spread and impact of this non-native invasive plant, and the efficacy of control techniques following a 3 year trial. Prepared on behalf of the New Forest Non-Native Plants Project, Freshwater Habitats Trust, Oxford, UK.

### 1.4.10 Use flame-throwers

- We found no evidence on the use of flame-throwers to control *Crassula helmsii*.

#### Background

The heat from flame-throwers can be used to destroy vegetation, and could therefore be used to control emergent *C. helmsii* vegetation. However, flame-

throwers may not produce enough heat to kill the roots and will not be effective on underwater plants, so would need to be used in combination with other approaches (Leach & Dawson 2000).

Leach J. & Dawson H. (2000) Is resistance futile? The battle against *Crassula helmsii*. *Journal of Practical Ecology and Conservation*, 4, 7-17.

### 1.4.11 Use dyes to reduce light levels

- One replicated, controlled study in the UK<sup>1</sup> found that applying aquatic dye, along with other treatments, did not reduce coverage of *Crassula helmsii*.

#### Background

Dye treatments work by reducing the amount of light that penetrates through water, and therefore reducing photosynthesis in plants under the water. This could potentially reduce the growth rate of *C. helmsii*. However, a study in the Netherlands suggested using soluble red and black dyes did not substantially reduce the amount of light penetrating the water. The authors concluded effective control was unlikely given the extreme adaptability of *C. helmsii* (EPPO 2014).

For studies covering the use of lightproof barriers to inhibit growth see 'Use lightproof barriers'. Increasing turbidity or using shading by vegetation to reduce growth are covered under the intervention 'Alter environmental conditions to control plants'.

EPPO (2014) PM 9/19 (1) Invasive alien aquatic plants. *EPPO Bulletin*, 44, 457-471.

A replicated, controlled study in 2011-2014 at waterbodies in the New Forest, UK (1) reported that treatment with aquatic dye, along with other treatments at some sites, did not reduce cover of *C. helmsii*, although no statistical tests were carried out. Average coverage of *C. helmsii* was 72% before and 75% at the end of the dye treatment, compared to 63% and 70% respectively at control sites. The study also showed that coverage of native plant species fell from 17% to 11% at treatment sites and from 17% to 14% at control sites over the trial period. Several other treatments (mechanical removal, herbicide, hot foam) were also used at some sites during this trial. A combination of Dyofix blue and black pond dyes were applied to six ponds on 5-6 occasions between August 2011 and December 2013. *C. helmsii* coverage was assessed in five random 0.25 m<sup>2</sup> quadrats within each treatment area in winter and summer from 2011-2014, and also in seven control ponds.

(1) Ewald N.C. (2014) *Crassula helmsii* in the New Forest. *Final report on the status, spread and impact of this non-native invasive plant, and the efficacy of control techniques following a 3 year trial*. Prepared on behalf of the New Forest Non-Native Plants Project, Freshwater Habitats Trust, Oxford, UK.

### 1.4.12 Use lightproof barriers

- Five before-and-after studies in the UK<sup>1,2,3,4,5</sup> found that covering *Crassula helmsii* with black sheeting or carpet strips eradicated<sup>1,2,3,4</sup> or severely reduced<sup>5</sup> the cover of the plant. However, *C. helmsii* was reported to have progressively recolonized two of the sites where it had been had initially been reported as eradicated<sup>2,3</sup>.

#### Background

Covering vegetation with lightproof barriers, such as matting or black sheeting, can control growth and eventually kill plants by preventing photosynthesis.

A before-and-after field trial in 2003 at waterbodies in a nature reserve in South Yorkshire, UK (1) reported that covering *C. helmsii* with black plastic and soil killed all plants, although no statistical tests were carried out. *C. helmsii* was covered with black plastic and topped with 1 m of soil in March 2003. No details about the area covered, duration of treatment or subsequent monitoring was provided.

A before-and-after field trial in 2003-2004 at a single pond in Bedfordshire, UK (2) reported that covering plants with black polythene eradicated *C. helmsii*, but it recolonized the site within a year and no statistical tests were carried out. Before the trial *C. helmsii* was estimated to cover 5% of the pond, and was eradicated after the treatment. However, one year after the treatment finished *C. helmsii* had recolonized the pond. The authors suggest this was due to plants which survived in surrounding areas not covered by the polythene. The 12 m<sup>2</sup> pond was covered with opaque black polythene weighed down with stones for six months between autumn 2003 and spring 2004.

A before-and-after study at a single lake in Dorset, UK (3) reported that covering *C. helmsii* with dark material killed the plant, although it slowly recolonized and no statistical tests were carried out. Two months after the dark sheeting was applied, the underlying *C. helmsii* was killed. However, after this the plant progressively recolonized the site. Typar geotextile sheeting was used to cover 50 m<sup>2</sup> of *C. helmsii*. No control or comparison, and few details of the experiment (e.g. timing, water depth), were provided.

A before-and-after study in 2000 at a lake in a nature reserve in Hampshire, UK (4) reported that covering with black sheeting killed *C. helmsii*, although no statistical tests were carried out. Black sheets (20 x 10 m) were secured tightly over submerged and exposed areas of *C. helmsii* for six months including summer. Few details of the site or methods were provided.

A before-and-after study in 2002-2004 at a single pond in Surrey, UK (5) reported that covering *C. helmsii* with carpet strips followed by the application of glyphosate reduced the area of the plant, although no statistical tests were carried out. One year after glyphosate treatment approximately 80% of *C. helmsii* had been killed, although it is not clear whether this was a direct result of the use

of carpet or was due to herbicide application. In autumn 2002, strips of carpet were placed over *C. helmsii* and weighted down wherever possible on the pond edge and in shallow water. In July-August 2003 the carpet was removed and the glyphosate-based herbicide Roundup was applied twice.

(1) Bridge T. (2005) Controlling New Zealand pygmyweed *Crassula helmsii* using hot foam, herbicide and by burying at Old Moor RSPB Reserve, South Yorkshire, England. *Conservation Evidence*, 2, 33-34.

(2) Wilton-Jones G. (2005) Control of New Zealand pygmyweed *Crassula helmsii* by covering with black polythene at The Lodge RSPB Reserve, Bedfordshire, England. *Conservation Evidence*, 2, 63.

(3) Dawson F. H. & Warman E.A. (1987) *Crassula helmsii* (T. Kirk) Cockayne: Is it an aggressive alien aquatic plant in Britain? *Biological Conservation*, 42, 247-272.

(4) Stone I. (2002) War against *Crassula* – one year on. *Enact*, 9-10.

(5) Anonymous (2004) Chemical control of Australian swamp stonecrop (New Zealand Pygmy Weed) *Crassula helmsii*. *The National Trust Conservation Newsletter*, 8, 2-3.

#### **1.4.13 Alter environmental conditions to control plants (e.g. shading by succession, increasing turbidity, re-profiling, or dredging)**

- No evidence was captured on altering environmental conditions to control *Crassula helmsii* by using shading, increasing turbidity, re-shaping or re-profiling banks of waterbodies or dredging.

#### **Background**

There are several possible methods of changing environmental conditions which could be used to reduce the growth of *C. helmsii*.

*C. helmsii* is relatively tolerant of shade, but heavy shade, such as that from overhanging willow trees could potentially reduce its competitive ability.

Increasing turbidity of the water to reduce light penetration and hence restrict plant growth has been suggested as a potential control method for *C. helmsii*. This could be achieved by releasing bottom-feeding fish, or seeding with nutrients to encourage algal growth (Leach & Dawson 2000).

Physical modification of waterbodies to increase the depth or make the sides steeper, could potentially reduce the area available for colonisation by *C. helmsii*. There is little evidence that dredging is an effective approach to control *C. helmsii*, although the total removal of the organic matter layer at the bottom of a pond was reportedly successful in eradicating *C. helmsii* from one site (Leach & Dawson 2000).

Leach J. & Dawson H. (2000) Is resistance futile? The battle against *Crassula helmsii*. *Journal of Practical Ecology and Conservation*, 4, 7-17.

#### **1.4.14 Plant other species to suppress growth of *Crassula helmsii***

- We found no evidence for the effects of using other plant species to control growth of *Crassula helmsii*.

##### **Background**

It has been suggested that the native species shoreweed *Littorella uniflora* can compete with *C. helmsii*, leading to reductions in cover of *C. helmsii* under some circumstances (Denton 2013). Denton (2013) suggests that there may be a chemical effect by which shoreweed inhibits the growth of *C. helmsii*.

Denton, J. (2013) Could shoreweed be useful for *Crassula* control? *Conservation Land Management*, 11, 18-19.

#### **1.4.15 Use grazing**

- One of two replicated, controlled studies in the UK found that excluding grazing reduced the abundance and coverage of *Crassula helmsii*<sup>1</sup>. The other study found no difference in cover of *C. helmsii* between ungrazed and grazed plots<sup>2</sup>.

##### **Background**

Grazing by livestock is often used as a conservation measure to increase plant species diversity in grassland. Depending on the dietary preferences of the animals, grazing could potentially increase or reduce the competitive advantage and coverage of *C. helmsii* on the margins of waterbodies or marshy areas. Increased grazing pressure was observed to be related to a decrease in *C. helmsii* cover at waterbodies in the New Forest, but several potentially confounding factors were present (Ewald 2014).

Ewald N.C. (2014) *Crassula helmsii* in the New Forest. Final report on the status, spread and impact of this non-native invasive plant, and the efficacy of control techniques following a 3 year trial. Prepared on behalf of the New Forest Non-Native Plants Project, Freshwater Habitats Trust, Oxford, UK.

A replicated, controlled study in 2012–2013 on the margins of a lake in Cambridgeshire, UK (1) found that excluding grazing reduced the abundance of *C. helmsii* compared to grazed plots. Cover of *C. helmsii* in ungrazed plots decreased from approximately 95% to 60% between July 2012 and October 2013, but remained above 90% in grazed plots. The abundance and diversity of other plants was higher in ungrazed compared to grazed plots (average abundance: 97% vs 38% cover respectively; mean species diversity (Shannon-Weiner): 1.1 vs 0.88). *C. helmsii* also

had lower proportional abundance in ungrazed compared to grazed plots (approximately 47% of total vegetation abundance vs 74%). Six 4 m<sup>2</sup> ungrazed fenced exclosures, interspersed with six 2 m<sup>2</sup> grazed plots, were set up in February 2012. The area was grazed by sheep in January-March 2012 and August 2012-October 2013, and by buffalo in July-December 2012. Percentage cover of *C. helmsii* and other plants was estimated eight times between July 2012 and October 2013.

A small, replicated, controlled study in 2009 at four ponds in the New Forest, UK (2) found that excluding grazing did not reduce the cover of *C. helmsii*. There was no significant difference between average cover of *C. helmsii* between ungrazed areas (42%) compared to grazed exclosures (26%). There was no difference in cover of plant species of conservation importance in ungrazed areas compared to grazed areas (7% vs 10%). Exclosure fences were erected in March 2009 to create ungrazed areas in four ponds with at least 75% *C. helmsii* cover. Grazing was mainly by ponies and cattle, but the area was also used by deer, pigs and donkeys. Exclosures included plants under the water and on the bank. Cover of plants in five random quadrats was surveyed in each pond in autumn 2009.

(1) Dean C.E., Day J., Gozlan R.E. & Diaz A. (2015) Grazing vertebrates promote invasive swamp stonecrop (*Crassula helmsii*) abundance. *Invasive Plant Science and Management*, 8, 131-138.

(2) Ewald N.C. (2014) *Crassula helmsii* in the New Forest. *Final report on the status, spread and impact of this non-native invasive plant, and the efficacy of control techniques following a 3 year trial*. Prepared on behalf of the New Forest Non-Native Plants Project, Freshwater Habitats Trust, Oxford, UK.

#### **1.4.16 Dry out waterbodies**

- We found no evidence for the effects of draining waterbodies on *Crassula helmsii*.

#### **Background**

Draining waterbodies is a potential tool for eliminating or reducing invasive aquatic organisms. One pond infested with *C. helmsii* was reported to have been drained over winter resulting in a reported 'severely reduced biomass' of the plant, although no data were provided (Dawson & Warman 1987). The ability of *C. helmsii* to withstand drying out (Anderson *et al.* 2015) and regenerate from small fragments of plant material (Dawson & Warman 1987) mean that this approach alone is unlikely to be fully effective. It is also likely to have negative impacts on native species.

Anderson L.G., Dunn A.M., Rosewarne P.J. & Stebbing P.D. (2015) Invaders in hot water: a simple decontamination method to prevent the accidental spread of aquatic invasive non-native species. *Biological Invasions*, 17, 2287-2297.

Dawson F. H. & Warman E.A. (1987) *Crassula helmsii* (T. Kirk) Cockayne: Is it an aggressive alien aquatic plant in Britain? *Biological Conservation*, 42, 247-272.

### 1.4.17 Bury plants

- We found no evidence on the use of burying with soil alone to control *Crassula helmsii*.

#### Background

Lightproof barriers have been shown to be effective in removing *C. helmsii* in some cases, and burying the plant under soil could be another potential approach. This could be carried out at the same time as creating a new pond, using the soil that is dug out to fill in a *C. helmsii*-infested water body. Studies investigating the use of burying with soil once a lightproof barrier has been put in place are discussed in 'Use lightproof barriers'.

### 1.4.18 Surround with wire mesh

- We found no evidence that surrounding *Crassula helmsii* with wire mesh reduced its rate of spread.

#### Background

*C. helmsii* can easily spread by regenerating from small fragments of plant material (Dawson & Warman 1987). Fine wire mesh (5 mm) can reduce the spread of *C. helmsii* by minimising movement of plant fragments to new areas. Mesh may need to fully surround the area of *C. helmsii*, including over the top of the affected area, to prevent spread by birds. This may be particularly important during physical disturbance caused by control treatments.

Dawson F. H. & Warman E.A. (1987) *Crassula helmsii* (T. Kirk) Cockayne: Is it an aggressive alien aquatic plant in Britain? *Biological Conservation*, 42, 247-272.

### 1.4.19 Decontamination to prevent further spread

- One controlled, replicated container study in the UK<sup>1</sup> found that submerging *Crassula helmsii* in hot water led to higher mortality than drying out plant fragments or a control.

#### Background

*C. helmsii* is able to rapidly regenerate from small fragments of plant, and is also highly tolerant of drying out (Dawson & Warman 1987). This means it can easily be spread between water bodies, for example on equipment used for fishing or other

watersports. Effective methods to decontaminate equipment, such as hot water or bleach, are therefore important in minimising the spread of *C. helmsii*.

Dawson F. H. & Warman E.A. (1987) *Crassula helmsii* (T. Kirk) Cockayne: Is it an aggressive alien aquatic plant in Britain? *Biological Conservation*, 42, 247-272.

A replicated, controlled container experiment in 2013-1014 in the UK (1) found that exposure to hot water led to higher mortality of *C. helmsii* fragments compared to drying treatment or a control. Submerging *C. helmsii* in hot water caused 90% mortality 1 h after treatment, and all plants were dead after 1 day. Hot water followed by drying did not result in additional mortality (80% mortality after 1 h). Drying treatment only led to partial mortality (20% after 8 days and 50% after 16 days), and all fragments in the control group survived for 16 days. Two hundred and forty 60 mm plant fragments were placed in mesh bags and submerged in 14 °C water for 1 h to simulate an angling trip. Hot water samples were then submerged in 45°C water for 15 min. Samples in the drying treatment were put on plastic trays in a room with circulating air. Control samples were placed in unsealed plastic bags to hinder drying. Mortality was assessed after 1 h and 1, 2, 4, 8 and 16 days using a FluorPen.

(1) Anderson L.G., Dunn A.M., Rosewarne P.J. & Stebbing P.D. (2015) Invaders in hot water: a simple decontamination method to prevent the accidental spread of aquatic invasive non-native species. *Biological Invasions*, 17, 2287-2297.

#### **1.4.20 Public education**

- No evidence was captured on the impact of education programmes on control of *Crassula helmsii*.

#### **Background**

*C. helmsii* can easily be dispersed between water bodies via a variety of management and recreational activities. Public education about the species and the need to decontaminate equipment could reduce the spread of *C. helmsii* from ornamental ponds and aquaria, and between natural water bodies. Direct sales of the species were banned in the UK in 2014.

#### **1.4.21 Use a combination of control methods**

- One before-and-after study at a single pond in the UK<sup>1</sup> found covering *Crassula helmsii* with carpet, followed by treatment with the herbicide glyphosate, killed 80% of the plant.

## Background

Because of the difficulty of controlling *C. helmsii*, a combination of control methods may be required to fully eradicate the species from a site (Dawson & Warman 1987, Leach & Dawson 2000, EPPO 2014). These could include a combination of mechanical removal, covering with lightproof barriers, and the use of herbicide. For example an integrated approach of mechanical removal (turf-stripping) and herbicide was used at a site in the UK, leading to an initial reduction in *C. helmsii*, although the species recolonized when treatment stopped and no quantitative data were provided (Clarke 2009).

Clarke S. (2009) *A summary of three different approaches to the treatment of non-native invasive species Crassula helmsii at protected sites*. Proceedings of the 41st Robson Meeting, 17-18 February 2009, Centre for Ecology and Hydrology, Wallingford, UK, 14-17.

Dawson F.H. & Warman E.A. (1987) *Crassula helmsii* (T. Kirk) cockayne: Is it an aggressive alien aquatic plant in Britain? *Biological Conservation*, 42, 247-272.

EPPO (2014) *PM 9/19 (1) Invasive alien aquatic plants*. EPPO Bulletin, 44, 457-471.

Leach J. & Dawson H. (2000) Is resistance futile? The battle against *Crassula helmsii*. *Journal of Practical Ecology and Conservation*, 4, 7-17.

A before-and-after study in 2002-2003 at a single pond in Surrey, UK (1) reported that covering *C. helmsii* with carpet strips followed by the application of glyphosate reduced the area of the plant, although no statistical tests were carried out. One year after glyphosate application approximately 80% of *C. helmsii* had been killed. In autumn 2002, strips of carpet were placed over *C. helmsii* and weighted down wherever possible on the pond edge and in shallow water. In July-August 2003 the carpet was removed and the glyphosate-based herbicide Roundup was applied twice.

(1) Anonymous (2004) Chemical control of Australian swamp stonecrop (New Zealand Pygmy Weed) *Crassula helmsii*. *The National Trust Conservation Newsletter*, 8, 2-3.

## 1.5 Parrot's feather *Myriophyllum aquaticum*

### Background

Parrot's feather *Myriophyllum aquaticum* (Vell.) Verdc., also known as Brazilian watermilfoil, is a perennial emergent freshwater plant native to tropical and subtropical South America. The species is a popular aquatic garden plant and has become a well-established invasive aquatic weed in numerous countries worldwide, including the USA, Japan, Australia, South Africa, Portugal, Italy and the UK (Hussner and Champion 2011).

In its native habitats, parrot's feather grows in warm, low altitude areas, in shallow waters and on muddy substrates. However, it can endure colder conditions and in its introduced range the species thrives in well-lit and still or slow-running waterbodies (CABI 2017). It normally grows rooted in shallow, nutrient-rich water, but it can also occur as a floating plant in deep water or as a terrestrial plant when ponds dry out (Fernandez *et al.* 1990; Cook 2004). This plasticity and its ability to reproduce through vegetative propagation (Nel *et al.* 2004), has facilitated the colonization of areas outside its native distribution. Although mostly benign in its indigenous range, in non-native areas it is often an aggressive competitor, capable of quick growth and spread (CABI 2017).

Parrot's feather plants can alter both the chemical and physical features of waterbodies and outside its native range the species has been found to reduce the diversity and abundance of native plants and to sometimes displace native species (EPPO 2004). Although it can offer cover and serve as food for some native aquatic organisms, when forming dense mats it can shade out algae and destabilise aquatic food chains (Global Invasive Species Database 2017). Dense concentrations of parrot's feather can clog waterways and irrigation channels, impairing navigation, the flow of irrigation water, fisheries and recreation (CABI 2017). Infestations can also affect hydroelectric power production (Fernandez *et al.* 1993) and facilitate mosquito habitat (Anderson 1993).

Early detection and rapid response is crucial for the control and eradication of invasive species. However, throughout much of its non-native range, the identification of parrot's feather is complicated by the occurrence of morphologically similar non-native and native *Myriophyllum* species (Hussner *et al.* 2017).

Anderson L.W.J. (1993). Aquatic weed problems and management in the western United States and Canada. Pages 371-391 in: A.H. Pieterse, K.J. Murphy (eds.) *Aquatic Weeds*. Oxford University Press, Oxford.

CABI (2017) *Myriophyllum aquaticum*. In: Invasive Species Compendium. Wallingford, UK: CAB International. Available at <http://www.cabi.org/isc/datasheet/34939> Accessed 2 October 2017.

Cook C.D.K (2004) *Aquatic and wetland plants of Southern Africa*. Backhuys Publishers, Leiden. Department of Ecology, State of Washington, (2009) *Non-native Invasive Freshwater Plants Parrotfeather (Myriophyllum aquaticum) Technical Information*. Available at <http://www.ecy.wa.gov/Programs/wq/plants/weeds/aqua003.html> Accessed 2 October 2017. EPPO (European and Mediterranean Plant Protection Organization) (2004) *Myriophyllum aquaticum*. EPPO data sheet on Invasive Plants *Myriophyllum aquaticum*. European and

Mediterranean Plant Protection. Available at [http://www.eppo.org/QUARANTINE/Pest\\_Risk\\_Analysis/PRAdocs\\_plants/draftds/05-11833%20DS%20Myriophyllum%20aquaticum.doc](http://www.eppo.org/QUARANTINE/Pest_Risk_Analysis/PRAdocs_plants/draftds/05-11833%20DS%20Myriophyllum%20aquaticum.doc) Accessed 2 October 2017.

Fernández O.A., Sutton D.L., Lallana V.H., Sabbatini M.R. & Irigoyen J. (1993) Aquatic weed problems and management in South and Central America. Pages 406-425 in: A.H. Pieterse, K.J. Murphy (eds.) *Aquatic Weeds*. Oxford University Press, Oxford.

Global Invasive Species Database (2017) *Myriophyllum aquaticum*. Available at <http://www.iucngisd.org/gisd/species.php?sc=401> on 26-09-2017 Accessed 2 October 2017.

Hussner A. & Champion P.D. (2011). *Myriophyllum aquaticum* (Vell.) verdcourt (parrot feather). Pages 103–112 in: Francis, R.A. (ed.) *A Handbook of Global Freshwater Invasive Species*. Earthscan Publisher, New York.

Hussner A., Stiers I., Verhofstad M.J.J.M., Bakker E.S., Grutters B.M.C., Haury J., van Valkenburg J.L.C.H., Brundu G., Newman J., Clayton J.S. & Anderson L.W.J. (2017) Management and control methods of invasive alien freshwater aquatic plants: A review. *Aquatic Botany*, 136, 112-137.

Nel J.L., Richardson D.M., Rouget M., Mgidi T.N., Mdzeke N., Maitre D.C. le, Wilgen B.W. van, Schonegevel L., Henderson L. & Naser S. (2004) A proposed classification of invasive alien plant species in South Africa: towards prioritizing species and areas for management action. *South African Journal of Science*, 100, 53-64.

## **Key messages**

### **1. Mechanical and physical control**

#### **1.1. Mechanical harvesting or cutting**

We captured no evidence for the effects of mechanical harvesting or cutting to control parrot's feather.

#### **1.2. Mechanical excavation**

We captured no evidence for the effects of mechanical excavation to control parrot's feather.

#### **1.3. Removal using water jets**

We captured no evidence for the effects of using water jets to control parrot's feather.

#### **1.4. Suction dredging and diver-assisted suction removal**

We captured no evidence for the effects of suction dredging or diver-assisted suction removal to control parrot's feather.

#### **1.5. Manual harvesting (hand-weeding)**

We captured no evidence for the effects of manual harvesting to control parrot's feather.

#### **1.6. Use of lightproof barriers**

We captured no evidence on the use of bottom shading to control parrot's feather.

#### **1.7. Water level drawdown**

One replicated, randomized, controlled laboratory study in the USA found that water removal to expose plants to drying during the summer led to lower survival of parrot's feather plants than water removal during winter.

#### **1.8. Dye application**

We captured no evidence for the effects of dye application to control parrot's feather.

### **2. Biological control**

#### **2.1. Biological control using fungal-based herbicides**

We captured no evidence for the effects of biological control of parrot's feather using fungal-based herbicides.

## **2.2. Biological control using herbivores**

Two replicated, randomized studies in Argentina and the USA found that stocking with grass carp reduced the biomass or abundance of parrot's feather. However, one controlled laboratory study in Portugal found that grass carp did not reduce biomass or cover of parrot's feather. One field study in South Africa found that one *Lysathia* beetle species retarded the growth of parrot's feather.

## **2.3. Biological control using plant pathogens**

One study in South Africa found that exposure to a strain of the bacterium *Xanthomonas campestris* did not affect the survival of parrot's feather.

# **3. Chemical control**

## **3.1. Use of herbicides**

### **3.1.1. Use of herbicides: 2,4-D**

Five laboratory studies (three replicated, controlled and two randomized, controlled) in the USA and Brazil and two replicated, randomized, field studies in Portugal reported that treatment with 2,4-D reduced growth, biomass or cover of parrot's feather.

### **3.1.2. Use of herbicides: carfentrazone-ethyl**

Five laboratory studies (one replicated, controlled, before-and-after, three replicated, controlled and one randomized, controlled) in the USA reported that treatment with carfentrazone-ethyl reduced growth.

### **3.1.3. Use of herbicides: diquat**

Two replicated, controlled laboratory studies in the USA reported reduced growth after exposure to diquat. However, one replicated, randomized, controlled field study in Portugal reported no reduction in biomass following treatment with diquat.

### **3.1.4. Use of herbicides: endothall**

Two replicated, controlled laboratory studies in the USA and New Zealand reported a reduction in biomass after treatment with endothall. However, one replicated, controlled field study in New Zealand found that cover declined after treatment with endothall but later cover increased close to pre-treatment levels.

### **3.1.5. Use of herbicides: triclopyr**

Three replicated, controlled laboratory studies in the USA and New Zealand reported that treatment with triclopyr reduced growth or that cover was lower than that of plants treated with glyphosate. One replicated, controlled field study and one replicated, before-and-after field study in New Zealand reported that cover was reduced after treatment with triclopyr but one of these studies reported that cover later increased to near pre-treatment levels.

### **3.1.6. Use of herbicides: other herbicides**

One replicated, randomized, controlled field study in Portugal and one replicated, controlled, laboratory study in the USA reported reduced growth or vegetation cover after treatment with glyphosate. Two replicated, randomized, controlled laboratory studies (one of which was randomized) in the USA have found that the herbicide imazapyr reduced growth. Four replicated, controlled (one of which was randomized) laboratory studies in the USA and New Zealand reported reduced growth after treatment with the herbicides imazamox, flumioxazin, dichlobenil and florpyrauxifen-benzyl. Two replicated, controlled (one of which was randomized) field studies in Portugal and New Zealand reported a decrease in cover after

treatment with dichlobenil followed by recovery. One replicated, randomized, controlled field study in Portugal reported reduced biomass after treatment with gluphosinate-ammonium. Three replicated, controlled laboratory studies in New Zealand and the USA found no reduction in growth after treatment with clopyralid, copper chelate or fluridone.

### **3.2. Use of salt**

We captured no evidence for the effects of treating parrot's feather with salt water.

## **4. Preventive management**

### **4.1. Decontamination / preventing further spread**

We captured no evidence on the effects of decontamination to prevent further spread of parrot's feather.

### **4.2. Public education**

We captured no evidence on the impact of education programmes on the control of parrot's feather.

### **4.3. Reduction of trade through legislation (e.g. trade ban)**

One randomized, before-and-after trial in the Netherlands reported that the implementation of a code of conduct reduced the trade of invasive aquatic plants banned from sale. One study in the USA found that despite a state-wide trade ban on parrot's feather plants, these could still be purchased in some stores.

## **5. Multiple integrated measures**

We captured no evidence on the use of multiple integrated measures to control parrot's feather.

### **1.5.1 Mechanical and physical control**

#### **1.5.1.1 Mechanical harvesting and cutting**

- We found no evidence on the use of manual harvesting to control parrot's feather.

#### **Background**

Mechanical control methods are widely used to control non-native and native weeds. Harvesting and cutting could be used to remove or reduce biomass of parrot's feather from affected waterbodies. Although the method is relatively cheap compared to other control options it is not species-specific and therefore may impact non-target taxa (Hussner *et al.* 2017). Due to its capacity to reproduce through vegetative propagation (CABI 2017), the use of this technique may be problematic for the control of parrot's feather as any fragments that remain may form new stands of vegetation. The use of manual harvesting to control parrot's feather is discussed in 'Manual harvesting (hand-weeding)'. The intervention 'Mechanical excavation' discusses the mechanical digging to control parrot's feather and the use of water-jet ventilation and suction dredging are respectively discussed in 'Removal using water jets' and 'Suction dredging and diver-assisted suction removal'.

CABI (2017) *Myriophyllum aquaticum*. In: Invasive Species Compendium. Wallingford, UK: CAB International. Available at <http://www.cabi.org/isc/datasheet/34939> Accessed 2 October 2017.

Hussner A., Stiers I., Verhofstad M.J.J.M., Bakker E.S., Grutters B.M.C., Haury J., van Valkenburg J.L.C.H., Brundu G., Newman J., Clayton J.S. & Anderson L.W.J. (2017) Management and control methods of invasive alien freshwater aquatic plants: A review. *Aquatic Botany*, 136, 112-137.

### **1.5.1.2 Mechanical excavation**

- We found no evidence on the use of mechanical excavation to control parrot's feather.

#### **Background**

Mechanical excavation can be used for digging sediment-rooted plants such as parrot's feather. Harvesting and digging using excavators, while not species-specific, can achieve considerable reduction of rooted floating-leaved, submerged and emergent plants in narrow waterbodies such as ponds, channels and small rivers (Hussner *et al.* 2017). The interventions 'Mechanical harvesting or cutting' discusses the mechanical harvesting or cutting of parrot's feather. The use of water-jet ventilation and suction dredging are respectively discussed in 'Removal using water jets' and 'Suction dredging and diver-assisted suction removal'. The use of manual harvesting to control parrot's feather is discussed in 'Manual harvesting (hand-weeding)'.

Hussner A., Stiers I., Verhofstad M.J.J.M., Bakker E.S., Grutters B.M.C., Haury J., van Valkenburg J.L.C.H., Brundu G., Newman J., Clayton J.S. & Anderson L.W.J. (2017) Management and control methods of invasive alien freshwater aquatic plants: A review. *Aquatic Botany*, 136, 112-137.

### **1.5.1.3 Removal using water jets**

- We found no evidence on the use of water jets to control parrot's feather.

#### **Background**

Water jets can be used to remove plants from sediments such as sand, peat and clay (Hussner *et al.* 2017). Plants are unrooted and can then be removed from the water surface. Compared to conventional mechanical harvesting methods, the use of water jets produces a lower number of plant fragments. However, as with other mechanic methods, the use of water jets to wash out plants leads to high water turbidity (Hussner *et al.* 2017). The control of parrot's feather by means of suction dredging is presented in 'Suction dredging and diver-assisted suction removal' and the use of manual and mechanical harvesting to control parrot's feather are respectively discussed in 'Manual harvesting (hand-weeding)' and 'Mechanical harvesting or

cutting'. Control via mechanical excavation is discussed under 'Mechanical excavation'.

Hussner A., Stiers I., Verhofstad M.J.J.M., Bakker E.S., Grutters B.M.C., Haury J., van Valkenburg J.L.C.H., Brundu G., Newman J., Clayton J.S. & Anderson L.W.J. (2017) Management and control methods of invasive alien freshwater aquatic plants: A review. *Aquatic Botany*, 136, 112-137.

#### **1.5.1.4 Suction dredging and diver-assisted suction removal**

- We found no evidence on the use of suction dredging and diver-assisted suction removal to control parrot's feather.

##### **Background**

Suction dredging consists of the use of high pressure water pumps to remove submerged vegetation. This method removes the plants with their root system and therefore reduces their capacity to regrow. The use of scuba divers allows for high species specificity and a combination of suction dredging and hand weeding has successfully eradicated small populations of other invasive aquatic plants (Hussner *et al.* 2017), including species of the same genus as parrot's feather (e.g. Eurasian watermilfoil *Myriophyllum spicatum*; Boylen *et al.* 1996). The control of parrot's feather using water jet ventilation is presented in 'Removal using water jets' and the use of manual and mechanical harvesting to control parrot's feather are respectively discussed in 'Manual harvesting (hand-weeding)' and 'Mechanical harvesting or cutting'. Control via mechanical excavation is discussed under 'Mechanical excavation'.

Boylen C.W., Eichler L.W., & Sutherland J.W. (1996). Physical control of Eurasian watermilfoil in an oligotrophic lake. *Hydrobiologia*, 340, 213-218.

Hussner A., Stiers I., Verhofstad M.J.J.M., Bakker E.S., Grutters B.M.C., Haury J., van Valkenburg J.L.C.H., Brundu G., Newman J., Clayton J.S. & Anderson L.W.J. (2017) Management and control methods of invasive alien freshwater aquatic plants: A review. *Aquatic Botany*, 136, 112-137.

#### **1.5.1.5 Manual harvesting (hand-weeding)**

- We found no evidence on the effects of manual to control parrot's feather.

##### **Background**

Hand-harvesting is a time-consuming yet highly species-specific method for the control of invasive aquatic plants. It has been used for the management of some problematic species (e.g. *Ludwigia peploides*; Husser *et al.* 2016); however, since parrot's feather is capable of reproducing through vegetative propagation this method might be problematic as any fragments that remain may form new stands of vegetation (Husser *et al.* 2017). Parrot's feather control using mechanical harvesting

is discussed under the intervention 'Mechanical harvesting or cutting' and the use of water jet ventilation and suction dredging are respectively discussed in 'Removal using water jets' and 'Suction dredging and diver-assisted suction removal'. 'Mechanical excavation' discusses the control of parrot's feather by means of mechanical digging.

Hussner A., Windhaus M. & Starfinger U. (2016) From weed biology to successful control: an example of successful management of *Ludwigia grandiflora* in Germany. *Weed Research*, 56, 434-441.

Hussner A., Stiers I., Verhofstad M.J.J.M., Bakker E.S., Grutters B.M.C., Haury J., van Valkenburg J.L.C.H., Brundu G., Newman J., Clayton J.S. & Anderson L.W.J. (2017) Management and control methods of invasive alien freshwater aquatic plants: A review. *Aquatic Botany*, 136, 112-137.

### **1.5.1.6 Use of lightproof barriers**

- We found no evidence on the use of lightproof barriers to control parrot's feather.

#### **Background**

Covering submerged weeds with lightproof barriers, such as plastic foils, tarpaulins or black sheeting, can control growth and eventually kill plants by preventing photosynthesis (De Winton et al. 2013). Lightproof barriers have been used to control other species of the same genus of parrot's feather with some success (Eurasian watermilfoil *Myriophyllum spicatum*) (Laitala et al. 2012). The method is not species-specific and its usage is usually restricted to small-scale management in slow-flowing or static waterbodies (Hussner et al. 2017).

De Winton M., Jones H., Edwards T., Özkundakci D., Wells R., McBride C., Rowe D., Hamilton D., Clayton J., Champion P. & Hofstra D. (2013) *Review of Best Management Practices for Aquatic Vegetation Control in Stormwater Ponds, Wetlands, and Lakes*. Auckland Council technical report, TR2013/026.

Hussner A., Stiers I., Verhofstad M.J.J.M., Bakker E.S., Grutters B.M.C., Haury J., van Valkenburg J.L.C.H., Brundu G., Newman J., Clayton J.S. & Anderson L.W.J. (2017) Management and control methods of invasive alien freshwater aquatic plants: A review. *Aquatic Botany*, 136, 112-137.

Laitala K.L., Prather T.S., Thill D., Kennedy B. & Caudill C. (2012). Efficacy of benthic barriers as a control measure for Eurasian watermilfoil (*Myriophyllum spicatum*). *Invasive Plant Science and Management* 5, 170-177

### **1.5.1.7 Water level drawdown**

- One replicated, randomized, controlled laboratory study in the USA<sup>1</sup> found that water removal to expose plants to drying during the summer led to lower survival of parrot's feather plants than exposing plants to drying during the winter.

## Background

This intervention, although limited to waterbodies in which water levels can be regulated, has the potential to reduce or eliminate aquatic invasive plants (Hussner *et al.* 2017). Water level drawdown consists of reducing the water level of a waterbody to expose submerged plants to drying (or freezing) conditions (De Winton *et al.* 2013). Desiccation may then result in plant mortality. However, parrot's feather plants can withstand some level of desiccation (Cook 2004) and therefore control using this technique is dependent on the duration of the water level drawdown. The reduction of water levels could also impact non-target native aquatic species.

Cook C.D.K (2004) *Aquatic and wetland plants of Southern Africa*. Backhuys Publishers, Leiden.

De Winton M., Jones H., Edwards T., Özkundakci D., Wells R., McBride C., Rowe D., Hamilton D., Clayton J., Champion P. & Hofstra D. (2013) *Review of Best Management Practices for Aquatic Vegetation Control in Stormwater Ponds, Wetlands, and Lakes*. Auckland Council technical report, TR2013/026.

Hussner A., Stiers I., Verhofstad M.J.J.M., Bakker E.S., Grutters B.M.C., Haury J., van Valkenburg J.L.C.H., Brundu G., Newman J., Clayton J.S. & Anderson L.W.J. (2017) Management and control methods of invasive alien freshwater aquatic plants: A review. *Aquatic Botany*, 136, 112-137.

A replicated, randomized, controlled, laboratory study conducted between 2008 and 2009 in the USA (1) found that water removal in order to expose plants to drying during the summer (summer dry outs) reduced survival of parrot's feather *Myriophyllum aquaticum* more than water removal during winter (winter dry outs). For four out of five comparisons, the survival of parrot's feather plants exposed to dry outs of the same duration was lower in summer (0–75%) than in winter (68–80%). After a dry out of 12 weeks, parrot's feather survival was 18% in summer and 78% in winter. Parrot's feather shoots were propagated in 3.78 l pots placed inside 1100 l containers filled with water. Four containers, each with 10 pots, were exposed to dry outs of two, four, six, eight and 12 weeks duration, or no dry out, in each season. Winter dry out was initiated in January and summer dry out was initiated in July.

(1) Wersal R.M., Madsen J.D. & Gerard P.D. (2013). Survival of parrotfeather following simulated drawdown events. *Journal of Aquatic Plant Management*, 51, 22-26.

### 1.5.1.8 Dye application

- We found no evidence on the use of dye treatments to control parrot's feather.

## Background

Dye treatments work by absorbing light that penetrates through water, and reducing the photosynthetic capacity of submerged plants (Hussner *et al.* 2017). However,

undesirable side effects have been reported for some *Myriophyllum* species following the use of dyes to control problematic epiphytes, leading to healthier and bigger *Myriophyllum* plants (Hussner *et al.* 2017). For studies covering the use of lightproof barriers to inhibit growth see 'Use of lightproof barriers'.

Hussner A., Stiers I., Verhofstad M.J.J.M., Bakker E.S., Grutters B.M.C., Haury J., van Valkenburg J.L.C.H., Brundu G., Newman J., Clayton J.S. & Anderson L.W.J. (2017) Management and control methods of invasive alien freshwater aquatic plants: A review. *Aquatic Botany*, 136, 112-137.

## 1.5.2 Biological control

### 1.5.2.1 Biological control using fungal-based herbicides

- We found no evidence for the effects of biological control of parrot's feather using fungal-based herbicides.

#### Background

Application of a mass-produced product, formulated from fungal microorganisms which cause disease in the host plant, can be used as a means of controlling invasive plants (Gassmann *et al.* 2006; Hussner *et al.* 2017). The use of pathogens to control parrot's feather is discussed under 'Biological control using plant pathogens'.

Gassmann A., Cock M.J.W., Shaw R. & Evans H.C. (2006) The potential for biological control of invasive alien aquatic weeds in Europe: a review. *Hydrobiologia*, 570, 217-222.

Hussner A., Stiers I., Verhofstad M.J.J.M., Bakker E.S., Grutters B.M.C., Haury J., van Valkenburg J.L.C.H., Brundu G., Newman J., Clayton J.S. & Anderson L.W.J. (2017) Management and control methods of invasive alien freshwater aquatic plants: A review. *Aquatic Botany*, 136, 112-137.

### 1.5.2.2 Biological control using herbivores

- One replicated, controlled laboratory study in Portugal<sup>1</sup> found that grass carp did not reduce biomass or cover of parrot's feather.
- Two replicated, randomized field studies in Argentina<sup>3</sup> and the USA<sup>4</sup> found that stocking with grass carp reduced the biomass<sup>3</sup> or abundance<sup>4</sup> of parrot's feather.
- One field study in South Africa<sup>2</sup> reported reduced growth of parrot's feather following the release a South American leaf-feeding *Lysathia* beetle.

#### Background

Both host-specific insects and grass carp have been used for the biocontrol of parrot's feather with some success (Moreira *et al.* 1999; Hill & Coetzee 2017), however, also native vertebrate herbivores and livestock can consume invasive

aquatic plants and consequently can contribute to inhibit their establishment, growth and expansion (Gassman et al. 2006).

The potential of host-specific insects to act as biocontrol agents depends on their ability to cause harm to the target plant, and may also be limited by the climatic conditions required by the insect species (Gassman et al. 2006). Although several insects have been suggested as potential control agents for parrot's feather (e.g. the stem-boring weevil *Listronotus marginicollis* has been found to show a feeding and host preference for parrot's feather, often killing its terminal bud (Oberholzer et al. 2007)), none seem to be in use. Grass carp *Ctenopharyngodon idella* consume large amounts of vegetation, and sterile fish have been used for the management and eradication of invasive aquatic plants (Hussner et al. 2017). However, grass carp are generalist herbivores and will consume all palatable plants available to them (Pine & Anderson 2001, Dorenbosch & Bakker 2011). The introduction of non-native control agents, such as herbivorous insects and grass carp, should only be considered following in-depth studies investigating possible undesired consequences to non-target species (Hussner et al. 2017).

Herbivory by both native vertebrates and livestock is often used as a conservation measure in terrestrial habitats. Depending on the dietary preferences of the animals, herbivory can also affect the competitive advantage of aquatic species such as parrot's feather, and consequently increase biotic resistance of freshwater ecosystems to invasive plants. As an example, herbivory by North American beavers *Castor canadensis* reduced the abundance of parrot's feather by 90% (Parker et al. 2007). The impact of terrestrial herbivores may be restricted to the margins of waterbodies or marshy areas, but aquatic and amphibious herbivores such as beavers can affect plant communities further away from water margins.

Dorenbosch M. & Bakker E.S. (2011). Herbivory in omnivorous fishes: effect of plant secondary metabolites and prey stoichiometry. *Freshwater Biology*, 56, 1783-1797.

Gassmann A., Cock M.J.W., Shaw R. & Evans H.C. (2006) The potential for biological control of invasive alien aquatic weeds in Europe: a review. *Hydrobiologia* 570, 217-222.

Hill, M. P., & Coetzee, J. (2017). The biological control of aquatic weeds in South Africa: Current status and future challenges. *Bothalia - African Biodiversity & Conservation*, 47, 1-12.

Hussner A., Stiers I., Verhofstad M.J.J.M., Bakker E.S., Grutters B.M.C., Haury J., van Valkenburg J.L.C.H., Brundu G., Newman J., Clayton J.S. & Anderson L.W.J. (2017) Management and control methods of invasive alien freshwater aquatic plants: A review. *Aquatic Botany*, 136, 112-137.

Moreira I., Ferreira T., Monteiro A., Catarino L. & Vasconcelos T. (1999) Aquatic weeds and their management in Portugal: insights and the international context. *Hydrobiologia*, 415, 229-234.

Oberholzer I.G., Mafokoane D.L. & Hill M.P. (2007) The biology and laboratory host range of the weevil, *Listronotus marginicollis* (Hustache)(Coleoptera: Curculionidae), a natural enemy of the invasive aquatic weed, parrot's feather, *Myriophyllum aquaticum* (Velloso) Verde (Haloragaceae). *African Entomology*, 15, 385-390.

Parker J.D., Caudill C.C., & Hay M.E. (2007). Beaver herbivory on aquatic plants. *Oecologia*, 151, 616-625.

Pine R.T. & Anderson L.W.J. (1991). Plant preferences of triploid grass carp. *Journal of Aquatic Plant Management* 29, 80-82.

A replicated, controlled, laboratory study from 1994 to 1996 in Portugal (1) found that grass carp *Ctenopharyngodon idella* did not reduce the biomass or cover of parrot's feather *Myriophyllum aquaticum*. Biomass and cover of parrot's feather did not differ between ponds where grass carp were present (biomass: 0.09 kg/m<sup>2</sup>,

cover: 5%) and ponds where carp were not present (0.09 kg/m<sup>2</sup> and 5%, respectively). Grass carp favoured soft-tissue native plants relative to parrot's feather. When presented only with parrot's feather and water hyacinth *Eichhornia crassipes*, one year old grass carp consumed parrot's feather at a daily rate of approximately 3% of their body weight but this increased to 20% by the age of two. Trials were conducted in six 660 l plastic tanks. Five tanks were stocked with carp and one control tank had no carp. Grass carp were presented with a selection of four plants with a total fresh weight similar to the biomass of fish present in the tank. After two days, the biomass of each plant species was weighed. The number of grass carp per tank was not specified.

A field study from 1995 to 1998 in a river in South Africa (2), reported reduced growth of parrot's feather *Myriophyllum aquaticum* following the release of a South American leaf-feeding *Lysathia* beetle. Three months after beetle release nearly all emergent parrot's feather shoots had been damaged by herbivory. After three years, 30% (558 out of 1251) of parrot's feather shoots were damaged by the *Lysathia* beetle. Damaged plants had lower mean shoot length (10 cm vs 19 cm) and dry weight (63 g vs 187 g/m<sup>2</sup>) compared to undamaged plants. Herbivory was reduced during winter. A total of 120 adult *Lysathia* beetles were released into one river site. Herbivory was quantified in ten 0.1 m<sup>2</sup> quadrats by counting the total number of shoots and the number of shoots with feeding damage. Sampling took place at intervals of four to six weeks for three years.

A replicated, randomized, controlled study from November 1996 to February 1997 in a water channel in Argentina (3) found that stocking with grass carp *Ctenopharyngodon idella* reduced the biomass of aquatic plants, including parrot's feather *Myriophyllum aquaticum*. After two months, dry weight of aquatic plants was lower in plots with grass carp at both low (50 g/m<sup>2</sup>) and high stocking densities (10 g/m<sup>2</sup>) than in plots without carp (320 g/m<sup>2</sup>). The experiment was performed in a medium size water channel with an aquatic plant community dominated by *Potamogeton pectinatus*, *M. aquaticum* and *Chara contraria*. Aquatic plant biomass was measured four times (sampling frequency not provided) in nine 30 m-long plots separated by iron barriers with plastic nets. Carp stocking density was 100 kg/ha (low density) and 200 kg/ha (high density).

A replicated, controlled, paired sites study from July 2005 to September 2007 in a reservoir in North Carolina, USA (4) found that high-density stocking with grass carp *Ctenopharyngodon idella* reduced the abundance of parrot's feather *Myriophyllum aquaticum*. For five out of six comparisons, the biomass of parrot's feather was lower in areas available for grass carp (0–113 g/m<sup>2</sup>) than in areas where grass carp were excluded (0–1330 g/m<sup>2</sup>). During the second year of the experiment no vegetation was detected in quadrats located in areas accessible to grass carp. Grass carp density was 100 fish/vegetated ha and grass carp were excluded from eight 6 m<sup>2</sup> areas using 1.3 cm plastic mesh. Vegetation in six 6 x 1 m quadrats was sampled monthly from July to September each year.

(1) Catarino L.F., Ferreira M.T. & Moreira, I.S. (1997). Preferences of grass carp for macrophytes in Iberian drainage channels. *Journal of Aquatic Plant Management*, 35, 79-83.

(2) Cilliers, C.J. (1999) *Lysathia* n. sp. (Coleoptera: Chrysomelidae), a host-specific beetle for the control of the aquatic weed *Myriophyllum aquaticum* (Haloragaceae) in South Africa. *Hydrobiologia*, 415, 271–276.

(3) Armellina, A.D., Bezic, C.R. & Gajardo, O.A. (1999). Submerged macrophyte control with herbivorous fish in irrigation channels of semiarid Argentina. *Hydrobiologia*, 415, 265–269.

(4) Garner A.B., Kwak T.J., Manuel K.L. & Barwick D.H. (2013). High-density grass carp stocking effects on a reservoir invasive plant and water quality. *Journal of Aquatic Plant Management*, 51, 27-33.

### **1.5.2.3 Biological control using plant pathogens**

- One study in South Africa<sup>1</sup> found that parrot's feather plants survived after being treated with a strain of the bacterium *Xanthomonas campestris*.

#### **Background**

Plant pathogens have been used to control invasive aquatic plants by inducing plant mortality (Charudattan 2001). The introduction of pathogens may impact non-target species and therefore in-depth studies investigating possible undesired consequences should be undertaken prior to the application of any plant pathogen. The use of fungal-based herbicides to control parrot's feather is discussed under 'Biological control using fungal-based herbicides' and the use of herbivores for the biocontrol of control parrot's feather is discussed under 'Biological control using herbivores'.

Charudattan, R. (2001). Biological control of weeds by means of plant pathogens: significance for integrated weed management in modern agro-ecology. *BioControl*, 46, 229-260.

A study in South Africa (1) reported that parrot's feather *Myriophyllum aquaticum* plants treated with a strain of the bacterium *Xanthomonas campestris* did not die. After treatment with a suspension of the bacterium all parrot's feather sections above the water died. However, after six weeks new shoots developed from the submerged stems leading to plant recovery. No data or statistics were reported. Plants were sprayed with a suspension of the bacterium at a concentration of 108 colony-forming units/ml. Authors do not report where or when the trials were conducted.

(1) Morris M.J., Wood A.R. & Den Breeÿen A. (1999). Plant pathogens and biological control of weeds in South Africa: a review of projects and progress during the last decade. *African Entomology Memoir*, 1, 129-137.

## **1.5.3 Chemical control**

### **1.5.3.1 Use of herbicides**

#### **1.5.3.1 Use of herbicides: 2,4-D**

- Three laboratory studies (including two replicated, controlled studies and one randomized, controlled study) in the USA<sup>5b, 9e</sup> and Brazil<sup>11</sup> found that the herbicide 2,4-D reduced the growth of parrot's feather.

- One replicated, controlled laboratory study in Brazil<sup>2</sup> found that 2,4-D led to a greater reduction in growth of parrot's feather than the herbicides diquat, glyphosate or imazapyr.
- One replicated, randomized, controlled field study in Portugal<sup>1c</sup> found that 2,4-D amine reduced the biomass of parrot's feather.
- One randomized, controlled field study in Portugal<sup>1e</sup> found that the combined application of 2,4-D and MCPA completely eliminated parrot's feather.
- One randomized, controlled laboratory study in the USA<sup>5c</sup> found that the combined application of 2,4-D and carfentrazone-ethyl led to a higher reduction in the cover of parrot's feather than the application of the herbicide dichlobenil eight days after treatment but not 45 days after treatment.

#### **1.5.3.2 Use of herbicides: carfentrazone-ethyl**

- Five laboratory studies (including one replicated, controlled, before-and-after study) in the USA<sup>4, 5a, 7b, 8b, 9f</sup> found that carfentrazone-ethyl reduced growth in parrot's feather.

#### **1.5.3.3 Use of herbicides: diquat**

- Two laboratory studies (including a replicated, randomized, controlled study) in the USA<sup>8a, 9b</sup> found that diquat reduced the growth of parrot's feather.
- One replicated, randomized, controlled field study in Portugal<sup>1d</sup> found that growth was not reduced after the application of diquat.

#### **1.5.3.4 Use of herbicides: endohall**

- Two replicated, controlled laboratory studies in New Zealand<sup>3a</sup> and the USA<sup>9c</sup> found that endohall reduced the growth of parrot's feather.
- One replicated, randomized, controlled field study in New Zealand<sup>3g</sup> found that parrot's feather plants treated with endohall presented lower cover soon after herbicide application but cover later increased to levels similar to pre-treatment.

#### **1.5.3.5 Use of herbicides: triclopyr**

- Two replicated, controlled laboratory studies in New Zealand<sup>3b</sup> and the USA<sup>9d</sup> reported reduced growth of parrot's feather following treatment with triclopyr.
- One replicated, before-and-after and one replicated, controlled field study in New Zealand<sup>3i, 3j</sup> found that cover was reduced after treatment with triclopyr. However, one of the studies noted that cover later increased to levels close to pre-treatment<sup>3i</sup>.
- One replicated, controlled laboratory study in New Zealand<sup>3f</sup> found that the application of triclopyr led to a greater reduction in cover than the application of glyphosate.

#### **1.5.3.6 Use of herbicides: other herbicides**

- One replicated, controlled laboratory study in New Zealand<sup>3c</sup> found that the application of dichlobenil reduced the growth of parrot's feather.
- Two replicated, randomized, controlled field studies in Portugal and New Zealand found that the application of dichlobenil reduced cover less than the combined application of the herbicides 2,4-D and MCPA eight days after treatment but not 45 days after treatment<sup>1f</sup> and that plants treated with dichlobenil presented lower

vegetation cover soon after herbicide application but cover later increased to levels close to pre-treatment<sup>3h</sup>.

- Three laboratory studies (including two replicated, controlled studies and one randomized, controlled study) in the USA found that the herbicides imazamox<sup>10b</sup> and imazapyr<sup>6, 10c</sup> reduced the growth of parrot's feather.
- One replicated, randomized, controlled field study in Portugal<sup>1a</sup> and one replicated, controlled, laboratory study in the USA<sup>10a</sup> reported reduced parrot's feather biomass after treatment with glyphosate. One replicated, randomized, controlled field study in Portugal<sup>1b</sup> found that the application of gluphosinate-ammonium reduced the biomass of parrot's feather.
- Three replicated, controlled laboratory studies in New Zealand and the USA found that treatment with fluridone<sup>3d</sup>, clopyralid<sup>3e</sup> and copper chelate<sup>9a</sup> did not reduce growth of parrot's feather.
- One replicated, controlled laboratory study in the USA<sup>7a</sup> found that the application of flumioxazin reduced the growth of parrot's feather.
- One replicated, randomized, controlled laboratory study in the USA<sup>12</sup> found that the application of florpyrauxifen-benzyl reduced the growth of parrot's feather.

## Background

Chemical herbicides have been used for the localized control of parrot's feather (e.g. Morreira *et al.* 1999). Herbicides licensed for use around the world use a wide diversity of different active ingredients (e.g. 2,4-D, imazapyr, carfentrazone-ethyl, diquat). However, herbicide control of aquatic vegetation is prohibited in numerous countries (e.g. no herbicides are approved for submerged species in Europe) (De Winton *et al.* 2013; Hussner *et al.* 2017). Therefore it is important to consider local regulations before using herbicide control. Legislative restrictions may be lifted for herbicides under particular conditions (Hussner *et al.* 2017). Impacts on non-target species should be considered prior to herbicide use.

Emerged and submerged vegetation may require different herbicides, and adjuvants may increase the efficacy of the treatment. More than one application of the herbicide is often required. The use of fungal-based herbicides to control parrot's feather is discussed under the intervention 'Biological control using fungal-based herbicides'

De Winton M., Jones H., Edwards T., Özkundakci D., Wells R., McBride C., Rowe D., Hamilton D., Clayton J., Champion P. & Hofstra D. (2013) *Review of Best Management Practices for Aquatic Vegetation Control in Stormwater Ponds, Wetlands, and Lakes*. Auckland Council technical report, TR2013/026.

Hussner A., Stiers I., Verhofstad M.J.J.M., Bakker E.S., Grutters B.M.C., Haury J., van Valkenburg J.L.C.H., Brundu G., Newman J., Clayton J.S. & Anderson L.W.J. (2017) Management and control methods of invasive alien freshwater aquatic plants: A review. *Aquatic Botany*, 136, 112-137.

Moreira I., Ferreira T., Monteiro A., Catarino L. & Vasconcelos T. (1999) Aquatic weeds and their management in Portugal: insights and the international context. *Hydrobiologia* 415, 229-234.

A replicated, randomized, controlled, field study conducted in summer 1986 in Portugal (1a), found that the application of the herbicide glyphosate reduced the

biomass of parrot's feather *Myriophyllum aquaticum*. For two out of three comparisons, the fresh weight of plants treated with glyphosate was reduced relative to untreated plants (9–14 vs 22–26 kg/m<sup>2</sup>). Additionally, four and a half months after treatment, the biomass of parrot's feather plants treated with glyphosate (13 kg/m<sup>2</sup>) was higher than plants treated with 2,4-D amine (2.2 kg/m<sup>2</sup>), but lower than plants treated with diquat (18 kg/m<sup>2</sup>). Parrot's feather biomass was assessed in 20 x 7 m plots, with four replicates of each herbicide. Herbicide rates were 2 kg/ha for diquat, 6.5 kg/ha for 2,4-D amine, 1–2.4 kg/ha for gluphosinate-ammonium and 3.6 kg/ha for glyphosate. Herbicide was applied twice in the same area.

A replicated, randomized, controlled, field study conducted in summer 1986 in three drainage channels Portugal (1b) found that the application of the herbicide gluphosinate-ammonium reduced the biomass of parrot's feather *Myriophyllum aquaticum* in five out of nine comparisons. For five out of nine comparisons, the fresh weight of parrot's feather plants treated with gluphosinate-ammonium was reduced relative to untreated plants (9–22 vs 22–26 kg/m<sup>2</sup>). Additionally, four and a half months after treatment, the biomass of parrot's feather plants treated with gluphosinate-ammonium (14–15 kg/m<sup>2</sup>) was higher than plants treated with 2,4-D amine (2.2 kg/m<sup>2</sup>) but lower than plants treated with glyphosate (13 kg/m<sup>2</sup>). Parrot's feather biomass was assessed in 20 x 7 m plots and each herbicide rate was tested in four replicates. Herbicide rates were 2 kg/ha for diquat, 6.5 kg/ha for 2,4-D amine, 1–2.4 kg/ha for gluphosinate-ammonium and 3.6 kg/ha for glyphosate. Herbicide was applied twice in the same area.

A replicated, randomized, controlled, field study conducted in summer 1986 in Portugal (1c) found that the application of the herbicide 2,4-D amine reduced the biomass of parrot's feather *Myriophyllum aquaticum*. For three out of three comparisons, the fresh weight of parrot's feather plants treated with 2,4-D amine was lower relative to untreated plants (2–9 vs 22–26 kg/m<sup>2</sup>). Additionally, four and a half months after treatment, the biomass of plants treated with 2,4-D amine (2.2 kg/m<sup>2</sup>) was significantly lower than that of plants treated with diquat (18 kg/m<sup>2</sup>), gluphosinate-ammonium (14–15 kg/m<sup>2</sup>) and glyphosate (13 kg/m<sup>2</sup>). Parrot's feather biomass was assessed in 20 x 7 m plots and each herbicide rate was tested in four replicates. Herbicide rates were 6.5 kg/ha for 2,4-D amine, 2 kg/ha for diquat, 1–2.4 kg/ha for gluphosinate-ammonium and 3.6 kg/ha for glyphosate.

A replicated, randomized, controlled, field study conducted in summer 1986 in Portugal (1d) found that the application of the herbicide diquat did not reduce the biomass of parrot's feather *Myriophyllum aquaticum*. For three out of three comparisons, the fresh weight of parrot's feather plants treated with diquat did not differ from untreated plants (15–25 vs 22–26 kg/m<sup>2</sup>). Additionally, the biomass of parrot's feather plants treated with diquat (15–25 kg/m<sup>2</sup>) was higher than that of plants treated with 2,4-D amine (2–9 kg/m<sup>2</sup>) for three out of three comparisons, was higher than that of plants treated with gluphosinate-ammonium (9–22 kg/m<sup>2</sup>) for five out of nine comparisons, and was higher than of plants treated with glyphosate (9–14 kg/m<sup>2</sup>) for two out of three comparisons. Parrot's feather biomass was assessed in 20 x 7 m plots and each herbicide rate was tested in four replicates. Herbicide rates were 2 kg/ha for diquat, 6.5 kg/ha for 2,4-D amine, 1–2.4 kg/ha for gluphosinate-ammonium and 3.6 kg/ha for glyphosate.

A replicated, controlled field study conducted in autumn 1995 in Portugal (1e) found that the combined application of the herbicides 2,4-D and MCPA led to a greater reduction of the cover of parrot's feather *Myriophyllum aquaticum* than the application of the herbicide dichlobenil eight days after treatment but not 45 days after application. Eight days after treatment, the cover of parrot's feather plants treated with a combination of 2,4-D and MCPA was lower than of plants treated with dichlobenil (10% vs 85%). However, after 45 days, the cover of plants treated with 2,4-D and MCPA was higher than that of plants treated with dichlobenil at a rate of 4.1 kg/ha (60% vs 20%). Each herbicide rate was sprayed onto three plots of 100 m<sup>2</sup>. Herbicide rates were 2.7 kg/ha and 4.1 kg/ha for dichlobenil and 520 g and 520 g/l for 2,4-D and MCPA respectively.

A replicated, controlled field study conducted in autumn 1995 in Portugal (1f) found that the application of the herbicide dichlobenil led to a smaller reduction in the cover of parrot's feather *Myriophyllum aquaticum* than the combined application of the herbicides 2,4-D and MCPA eight days after treatment but not 45 days after application. Eight days after treatment, the cover of parrot's feather plants treated with dichlobenil (85% cover) was higher than of plants treated with a combination of 2,4-D and MCPA (10%). However, after 45 days, the cover of plants treated with dichlobenil at a rate of 4.1 kg/ha (20%) was lower than of plants treated with a dichlobenil at a rate of 2.7 kg/ha (60%) or with a combination of 2,4-D and MCPA (60%). Each herbicide rate was sprayed onto three plots of 100 m<sup>2</sup>. Herbicide concentration was 520 g and 520 g/l for 2,4-D + MCPA respectively.

A small, replicated, controlled, laboratory study conducted in 1999 in Brazil (2) found that the application of the herbicide 2,4-D above a certain concentration led to a greater reduction in growth in parrot's feather *Myriophyllum aquaticum* than the application of the herbicides diquat, glyphosate or imazapyr. Thirty-six days after application, control of parrot's feather plants by 2,4-D (4–100%, visual assessment with 0% corresponding to no control and 100% to complete control) was higher than control by diquat (53–54%) in 11 out of 12 comparisons, control by glyphosate (34%), and control by imazapyr (8.5%). However, the application of 2,4-D at a concentration of 167 g/ha led only to 4% control. Fifteen parrot's feather shoots were propagated in 120 l containers filled with water. Herbicide concentrations tested were 167, 335, 670 and 1340 g/ha for 2,4-D, 102 and 204 g/ha for diquat, 3360 g/ha for glyphosate and 250 g/ha for imazapyr. Control in the context of the visual assessments is not clearly defined.

A small, replicated, controlled, laboratory study conducted between 1999 and 2000 in New Zealand (3a) found that the herbicide endothall reduced the growth of parrot's feather *Myriophyllum aquaticum*. After 17 weeks, plants treated with endothall had a lower dry weight (29–57 g) than that of untreated plants (274 g). Plants were grown for approximately two months prior to herbicide application in 60 l plastic tubs. Endothall was sprayed onto plants in three tubs at a concentration of 9 and 15 kg/ha and plants in four tubs were left untreated.

A small, replicated, controlled, laboratory study conducted between 1999 and 2000 in New Zealand (3b) found that the herbicide triclopyr reduced the growth of parrot's feather *Myriophyllum aquaticum*. After 17 weeks, plants treated with triclopyr had a lower dry weight (1–2 g) than that of untreated plants (274 g). Plants were grown for approximately two months prior to herbicide application in 60 l

plastic tubs. Triclopyr was sprayed onto plants in three tubs at a concentration of 2 and 4 kg/ha and plants in four tubs were left untreated.

A small, replicated, controlled, laboratory study conducted between 1999 and 2000 in New Zealand (3c) found that the herbicide dichlobenil reduced the growth of parrot's feather *Myriophyllum aquaticum*. After 17 weeks, laboratory plants treated with dichlobenil had a lower dry weight (6–21 g) than that of untreated plants (274 g). Plants were grown for approximately two months prior to herbicide application in 60 l plastic tubs. Dichlobenil was sprayed onto plants in three tubs at a concentration of 2 and 4 kg/ha and plants in four tubs were left untreated.

A small, replicated, controlled, laboratory study conducted between 1999 and 2000 in New Zealand (3d) found that the biomass of parrot's feather *Myriophyllum aquaticum* treated with the herbicide fluridone did not differ significantly from that of untreated plants. After 17 weeks, the dry weight of laboratory plants treated with fluridone (176–216 g) was not significantly different from that of untreated plants (274 g). Plants were grown for approximately two months prior to herbicide application in 60 l plastic tubs. Fluridone was sprayed onto plants in three tubs at a concentration of 0.1 and 0.5 kg/ha and plants in four tubs were left untreated.

A small, replicated, controlled, laboratory study conducted between 1999 and 2000 in New Zealand (3e) found that the biomass of parrot's feather *Myriophyllum aquaticum* treated with the herbicide clopyralid did not differ significantly from that of untreated plants. After 17 weeks, the dry weight of plants treated with clopyralid (132 g) was not significantly different from that of untreated plants (274 g). Plants were grown for approximately two months prior to herbicide application in 60 l plastic tubs. Clopyralid was sprayed onto plants in three tubs at a concentration of 1.5 kg/ha and plants in four tubs were left untreated.

A small, replicated, controlled, laboratory study conducted between 1999 and 2000 in New Zealand (3f) found that the vegetation cover of parrot's feather *Myriophyllum aquaticum* plants treated with triclopyr was lower than that of plants treated with the glyphosate. One year after exposure, the vegetation cover of plants treated with triclopyr ranged between 0 and 13% whereas the vegetation cover of plants treated with glyphosate was 73%. The percentage cover of untreated plants was 83%. Plants were grown for approximately two months prior to herbicide application in 60 l plastic tubs. Triclopyr was applied at concentrations of 2, 4 and 8 kg/ha whereas glyphosate was sprayed onto plants in four tubs at a concentration of 3.2 kg/ha and plants in four tubs were left untreated.

A replicated, controlled field study conducted between 2001 and 2002 in a wetland in the Northern Island of New Zealand (3g) reported that treatment with the herbicide endothall reduced vegetation cover of parrot's feather *Myriophyllum aquaticum* plants soon after application, but after 28 weeks cover was similar to pre-treatment levels. Results were not subject to statistical tests. After 10 weeks and following a second herbicide application, vegetation cover of treated plants was lower (2%) than untreated plants (47%). However, after 28 weeks, vegetation cover of treated plants (93%) was similar to that of untreated plants (97%). Authors reported that the increase in vegetation cover resulted from the encroachment of plants from outside sprayed areas rather than due to regrowth in treated plots. Endothall was applied at concentrations of 8.8 and 14.8 kg/ha. Each herbicide concentration was sprayed into three 5 x 5 m plots and three plots were left

untreated. Herbicides were applied in early summer (December). A second application took place 51 days after the initial treatment.

A replicated, controlled field study conducted between 2001 and 2002 in a wetland in the Northern Island of New Zealand (3h) reported treatment with the herbicide dichlobenil reduced vegetation cover of parrot's feather *Myriophyllum aquaticum* plants soon after application, but after 28 weeks cover was similar to pre-treatment levels. Results were not subject to statistical tests. After 10 weeks and following a second herbicide application, vegetation cover of treated plants was lower (3–8%) than untreated plants (47%). However, after 28 weeks, vegetation cover of treated plants (70–98%) was similar to that of untreated plants (97%). Authors reported that the increase in vegetation cover resulted from the encroachment of plants from outside sprayed areas rather than due to regrowth in treated plots. Dichlobenil was applied at concentrations of 6.8 and 20.3 kg/ha. Each herbicide concentration was sprayed into three 5 x 5 m plots and three plots were left untreated. Herbicides were applied in early summer (December). A second application took place 51 days after the initial treatment.

A replicated, controlled field study conducted between 2001 and 2002 in a wetland in the Northern Island of New Zealand (3i) reported treatment with the herbicide triclopyr reduced vegetation cover of parrot's feather *Myriophyllum aquaticum* plants soon after application, but after 28 weeks cover was similar to pre-treatment levels. Results were not subject to statistical tests. After 10 weeks and following a second herbicide application, vegetation cover of treated plants was lower (1.5%) than of untreated plants (47%). However, after 28 weeks, vegetation cover of treated plants (68–84%) was similar to that of untreated plants (97%). Authors reported that the increase in vegetation cover resulted from the encroachment of plants from outside sprayed areas rather than due to regrowth in treated plots. Triclopyr was applied at concentrations of 2 and 4 kg/ha. Each herbicide concentration was sprayed into three 5 x 5 m plots and three plots were left untreated. Herbicides were applied in early summer (December). A second application took place 51 days after the initial treatment.

A replicated, before-and-after field study conducted between 2002 and 2003 in two drains in the Northern Island of New Zealand (3j) reported a reduction in the cover of parrot's feather *Myriophyllum aquaticum* after the application of the herbicide triclopyr. This result is not based on statistical tests. The areas occupied by parrot's feather were greater before herbicide application (35 m<sup>2</sup> and 128 m<sup>2</sup>) than following herbicide application (22 m<sup>2</sup> and 2 m<sup>2</sup>, respectively). Authors reported that native species such as *Potamogeton cheesemanii* and *Persicaria decipiens* were either not affected or recovered quickly, although no data were presented. Triclopyr was applied at a concentration of 4 kg/ha into two sections with a low density of parrot's feather (one 3 m x 2 km and the other 1 km long, being 3 m wide in the first 500 m and 1 m wide in the remaining 500 m). Application occurred during the spring and summer of 2002 to 2003 and vegetation cover was assessed visually.

A small, replicated, controlled, before-and-after, laboratory study conducted in 2003 in the USA (4), found that the application of the herbicide carfentrazone-ethyl reduced growth in parrot's feather *Myriophyllum aquaticum* but did not lead to plant death. After 28 days, the biomass of parrot's feather shoots treated with carfentrazone-ethyl was 29.5–54% lower than untreated plants. However, emerged

and submerged foliage remained viable. The study was carried out using 12 1,600 l containers filled with water, each of which contained eight 5 l containers with three 15 cm parrot's feather stems. Each of the three tested herbicide rates (50, 100 and 200 µg/l) was applied to three 1600 l containers, and plants in three containers were left untreated. Plants from four of the eight 5 l plastic containers placed inside each 1,600 l container were harvested, dried and weighed before herbicide application, and the remaining plants were harvested, dried and weighed 28 days after herbicide application.

A small, replicated, randomized, controlled, laboratory study conducted in 2004 in the USA (5a), found that the herbicide carfentrazone-ethyl reduced growth in parrot's feather *Myriophyllum aquaticum*. After three weeks, young parrot's feather plants treated with carfentrazone-ethyl had a lower dry weight (1.8–2.4 g/pot) than untreated plants (6.4–10 g/pot). Parrot's feather shoots were propagated in 3.9 l plastic containers. Carfentrazone-ethyl application rate ranged between 100 and 200 µg/l and each herbicide rate was applied to three plants.

A small, replicated, randomized, controlled, laboratory study conducted in 2004 in the USA (5b), found that the herbicide 2,4-D reduced growth in parrot's feather *Myriophyllum aquaticum*. After three weeks, young parrot's feather plants treated with 2,4-D had a lower dry weight (0–3.1 g/pot) than untreated plants (6.4–10 g/pot). Parrot's feather shoots were propagated in 3.9 l plastic containers. 2,4-D application rate ranged between 100 and 1,000 µg/l and each herbicide rate was applied to three plants.

A small, replicated, randomized, controlled, laboratory study conducted in 2004 in the USA (5c), found that the combined application of the herbicides 2,4-D and carfentrazone-ethyl completely eliminated parrot's feather *Myriophyllum aquaticum*. After three weeks, young parrot's feather plants treated with a combination of 2,4-D and carfentrazone-ethyl were totally controlled (0 g/pot) whereas untreated plants had a biomass between 6.4 and 10 g/plot. Parrot's feather shoots were propagated in 3.9 l plastic containers. Carfentrazone-ethyl was applied at a constant rate of 100 µg/l, whereas the rate of 2,4-D application ranged between 250 and 2,000 µg/l. Each herbicide rate was applied to three plants.

A small, replicated, randomized, controlled, laboratory study conducted in 2006 in the USA (6) found that the application of the herbicide imazapyr reduced growth in parrot's feather *Myriophyllum aquaticum*. After ten weeks, the biomass of parrot's feather plants treated with imazapyr at rates of 584 and 1,123 g/ha was reduced to 0 g/pot whereas untreated plants had a biomass of 140 g/pot. The biomass of plants treated with imazapyr at a rate of 281 g/ha (160 g/pot) did not differ significantly from untreated plants (140 g/pot). Parrot's feather shoots were propagated in 3.8 l plastic containers. Each herbicide rate (281, 584 and 1123 g/ha) was applied to three plants and three plants were left untreated.

A small, replicated, controlled, laboratory study conducted in the USA (7a), found that the herbicide flumioxazin reduced growth in parrot's feather *Myriophyllum aquaticum*. Parrot's feather plants sprayed with flumioxazin had a lower dry weight (0.29–0.74 g) than unsprayed plants (1.43 g). Parrot's feather shoot tips (5–10 cm) were transplanted into 9 cm<sup>2</sup> pots. Application rate of the herbicide flumioxazin ranged between 34 and 437 g/ha and each herbicide rate was applied to pots with

three plants and plants in one pot were left unsprayed. Dates of the studies are not presented.

A small, replicated, controlled, laboratory study conducted in the USA (7b), found that the herbicide carfentrazone-ethyl reduced growth in parrot's feather *Myriophyllum aquaticum*. Parrot's feather plants treated with carfentrazone-ethyl had a lower dry weight (0.55–0.88 g) than untreated plants (1.43 g). Parrot's feather shoot tips (5–10 cm) were transplanted into 9 cm<sup>2</sup> pots. Carfentrazone-ethyl application rate ranged between 56 and 224 g/ha and each herbicide rate was applied to pots with three plants and plants in one pot were left unsprayed. Dates of the studies are not presented.

A small, replicated, randomized, controlled, laboratory study conducted in 2006 in the USA (8a), found that the application of the herbicide diquat reduced growth of parrot's feather *Myriophyllum aquaticum*. After four weeks, the dry weight of parrot's feather plants treated with diquat was lower than untreated plants (1–3 vs 6 g/pot). Daytime and night-time application of the herbicide resulted in similar results. Parrot's feather shoots were propagated in 3.78 l pots and placed inside 246 l containers filled with water. Each herbicide rate (0.19 and 0.37 mg/l) was applied to three plants.

A small, replicated, randomized, controlled, laboratory study conducted in 2006 in the USA (8b), found that the application of the herbicide carfentrazone-ethyl reduced growth in parrot's feather *Myriophyllum aquaticum*. After four weeks, the dry weight of parrot's feather plants treated with carfentrazone-ethyl was lower than untreated plants (1–2.1 vs 4.6 g/pot). Daytime and night-time application of the herbicide resulted in similar results. Parrot's feather shoots were propagated in 3.78 l pots and placed inside 246 l containers filled with water. Each herbicide rate (0.1 and 0.2 mg/l) was applied to three plants.

A small, replicated, controlled, laboratory study conducted between 2007 and 2008 in the USA (9a) found that the application of the herbicide copper chelate did not affect the growth of parrot's feather *Myriophyllum aquaticum*. After six weeks, the dry weight of parrot's feather treated with copper chelate (14–16 g/pot) did not differ significantly from the biomass of untreated plants (18 g/pot). Visual assessment revealed no reduction in plant vegetation by copper chelate compared to untreated controls six weeks after herbicide application. Parrot's feather shoots were propagated in 3.78 l pots and placed inside 246 l containers filled with water. Each herbicide rate (0.5 and 1 mg/l) was applied to four 246 l containers, each holding four plants. Number of plants used as control is not presented and control in the context of the visual assessments was not clearly defined.

A small, replicated, controlled, laboratory study conducted between 2007 and 2008 in the USA (9b), found that the application of the herbicide diquat reduced growth in parrot's feather *Myriophyllum aquaticum*. After six weeks, the dry weight of parrot's feather plants treated with diquat was reduced compared to untreated plants (2–6 vs 18 g/pot). Six weeks after application, diquat had controlled parrot's feather plants by 50–70% (visual assessment, with 0% corresponding to no control and 100% to complete control). Parrot's feather shoots were propagated in 3.78 l pots and placed inside 246 l containers filled with water. Each herbicide rate (subsurface: 0.19 and 0.37 mg/l; foliar: 4.5 kg/ha) was applied to four 246 l containers, each holding four plants. Number of plants used as control is not

presented and control in the context of the visual assessments was not clearly defined.

A small, replicated, controlled, laboratory study conducted between 2007 and 2008 in the USA (9c), found that the application of the herbicide endothall above a certain concentration reduced the growth of parrot's feather *Myriophyllum aquaticum*. After six weeks, the dry weight of parrot's feather plants treated with endothall at a concentration of 5 mg/l was lower than that of untreated plants (12 vs 18 g/pot) but the dry weight of plants treated with endothall at a concentration of 2.5 mg/l did not differ from untreated plants (17 vs 18 g/pot). Visual assessment revealed no reduction in vegetation by endothall at either concentration six weeks after herbicide application (0% change relative to untreated plants). Parrot's feather shoots were propagated in 3.78 l pots and placed inside 246 l containers filled with water. Each herbicide rate was applied to four 246 l containers, each holding four plants. Number of plants used as control is not presented. Visual assessments were expressed in percentage, with 0% corresponding to no control and 100% to complete control.

A small, replicated, controlled, laboratory study conducted between 2007 and 2008 in the USA (9d), found that the application of the herbicide triclopyr reduced growth in parrot's feather *Myriophyllum aquaticum*. After six weeks, the dry weight of parrot's feather plants treated with triclopyr was lower than that of untreated plants (3–6 vs 10 g/pot). Subsurface and foliar herbicide applications led to similar changes in biomass. Six weeks after application, triclopyr had controlled parrot's feather by 15–70% (visual assessment with 0% corresponding to no control and 100% to complete control). Parrot's feather shoots were propagated in 3.78 l pots and placed inside 246 l containers filled with water. Each herbicide rate (subsurface: 1.25 and 2.5 mg/l; foliar: 6.7 kg/ha) was applied to four 246 l containers, each holding four plants. Number of plants used as control is not presented and control in the context of the visual assessments was not clearly defined.

A small, replicated, controlled, laboratory study conducted between 2007 and 2008 in the USA (9e), found that the application of the herbicide 2,4-D above a certain concentration reduced growth in parrot's feather *Myriophyllum aquaticum*. After six weeks, the dry weight of plants treated with subsurface 2,4-D at a concentration of 5 mg/l was lower than that of untreated plants (10 vs 18 g/pot). However, the dry weight of plants treated with 2,4-D at a concentration of 2 mg/l did not differ from that of untreated plants (15 vs 18 g/pot). Dry weight of plants exposed to foliar application of 2,4-D (1 g/pot) was lower than untreated plants or those treated with 2,4-D underwater. Parrot's feather shoots were propagated in 3.78 l pots and placed inside 246 l containers filled with water. Each herbicide rate was applied to four 246 l containers, each holding four plants. Number of plants used as control is not presented.

A small, replicated, controlled, laboratory study conducted between 2007 and 2008 in the USA (9f) found that the application of the herbicide carfentrazone-ethyl reduced growth of parrot's feather *Myriophyllum aquaticum*. After six weeks, the dry weight of parrot's feather plants treated with carfentrazone-ethyl was lower (10–12 g/pot) than that of untreated plants (18 g/pot). Six weeks after application, plants treated with carfentrazone-ethyl were reduced by 0–15% (visual assessment with 0% corresponding to no reduction in cover relative to untreated plans and 100% to

complete elimination). Parrot's feather shoots were propagated in 3.78 l pots and placed inside 246 l containers filled with water. Each herbicide rate (0.1 and 0.2 mg/l) was applied to four 246 l containers, each holding four plants. Number of plants used as control is not presented.

A small, replicated, controlled, laboratory study conducted in the USA (10a) found that the herbicide glyphosate reduced the growth of parrot's feather *Myriophyllum aquaticum*. After five weeks, plants treated with glyphosate had a lower dry weight (0.49 g) than untreated plants (3.15 g). Application of glyphosate to parrot's feather regrowth led to a similar biomass reduction. Dry weight of plants treated with glyphosate did not differ from that of plants treated with the herbicide imazamox (0.97 g) or imazapyr (0.39 g). The plants were grown for approximately three weeks prior to herbicide application and each treatment had four replicates. Herbicides were sprayed on plants with no submerged vegetation at a concentration of 2240 g/ha for glyphosate and 560 g/ha for both imazamox and imazapyr.

A small, replicated, controlled, laboratory study conducted in the USA (10b) found that the herbicide imazamox reduced the growth parrot's feather *Myriophyllum aquaticum*. After five weeks, plants treated with imazamox had a dry weight (0.97 g) approximately 40–85% lower than that of untreated plants (3.15 g). Application of imazamox to parrot's feather regrowth led to similar biomass reduction. Dry weight of plants treated with imazamox did not differ from the dry weight of plants treated with the herbicides glyphosate (0.49 g) or imazapyr (0.39 g). The plants were grown for approximately three weeks prior to herbicide application and each treatment had four replicates. Herbicides were sprayed on plants with no submerged growth. Concentration of imazamox ranged from 35 to 580 g/ha, glyphosate was applied at 2240 g/ha and imazapyr at 560t g/ha.

A small, replicated, controlled, laboratory study conducted in the USA (10c) found that the herbicide imazapyr reduced the growth of parrot's feather *Myriophyllum aquaticum*. After five weeks, plants treated with imazapyr had a lower dry weight (0.39 g) than that of untreated plants (3.15 g). Application of imazapyr to parrot's feather regrowth led to similar biomass reduction. Dry weight of plants treated with imazapyr did not differ from the dry weight of plants treated with the herbicide imazamox (0.97 g) or glyphosate (0.49 g). The plants were grown for approximately three weeks prior to herbicide application and each treatment had four replicates. Herbicides were sprayed on plants with no submersed growth. Herbicide concentration was 560 g/ha for both imazapyr and imazamox and 2240 g/ha for glyphosate.

A small, replicated, controlled, laboratory study conducted in Brazil (11), found that the application of the herbicide 2,4-D reduced plant growth in parrot's feather *Myriophyllum aquaticum* even when plants were exposed to simulated rainfall. Seven days after the application of 2,4-D at concentrations of 670 g/ha and 1340 g/ha, treated parrot's feather plants were controlled by 61.5% and 81.5% respectively, when exposed to simulated rain 15 minutes after herbicide application. Exposure to simulated rain only influenced treatment if it occurred within 15 minutes of herbicide application and for a 2,4-D concentration of 670 g/ha. With or without simulated rain, the control was almost complete 21 days after the application of 2,4-D (commercial version DMA 806 BR). Parrot's feather shoots (20

cm) were transplanted into plastic containers and each treatment had four replicates. The metric used to define control is not clearly presented.

A small, replicated, randomized, controlled laboratory study conducted in the USA (12), found that a new herbicide tested under the code SX1552 (chemical name: 4-amino-3-chloro-6-(4-chloro-2-fluoro-3-methoxyphenyl)-5-fluoro-pyridine-2-benzyl ester; common name florpiauxifen-benzyl) reduced parrot's feather *Myriophyllum aquaticum* growth. Four weeks after exposure, the dry weight of parrot's feather plants treated with SX1552 was reduced to less than 80% of the dry weight of untreated plants (data not reported). Experiments were based on laboratory stock plants grown in 15 l plastic containers. Herbicide concentration ranged between 3 and 81 µg/l. Each SX1552 concentration was tested in four plants. Plants were grown for about one week prior to treatment.

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### 1.5.3.1 Use of salt

- We found no evidence on the impact of using salt on the control of parrot's feather.

#### Background

Salt has traditionally been used as a non-selective herbicide for controlling problematic aquatic weeds and the addition of salt has been suggested as a potential

control method for parrot's feather (Hussner et al. 2017). However, parrot's feather appear to have some resistance to salt stress and may even colonize brackish water (Thouvenot et al, 2012).

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## **1.5.4 Preventive management**

### **1.5.4.1 Decontamination / preventing further spread**

- We found no evidence on the effects of decontamination to prevent further spread of parrot's feather.

#### **Background**

Parrot's feather plants are capable of rapidly regenerating from small fragments, and are highly tolerant of desiccation (Hussner & Champion 2011). As a consequence the species can be easily transported between waterbodies when attached to fishing or watersport equipment, imported with soil or as fragments on machinery. The decontamination of equipment and matter, using for instance hot water or bleach, could therefore potentially limit the spread of the species.

Hussner A., Stiers I., Verhofstad M.J.J.M., Bakker E.S., Grutters B.M.C., Haury J., van Valkenburg J.L.C.H., Brundu G., Newman J., Clayton J.S. & Anderson L.W.J. (2017) Management and control methods of invasive alien freshwater aquatic plants: A review. *Aquatic Botany*, 136, 112-137.

### **1.5.4.2 Public education**

- We found no evidence on the impact of education programmes on the control of parrot's feather.

#### **Background**

Public education about parrot's feather and its impact on the ecosystems where it is introduced can potential change people's behaviour, reducing its use in ornamental ponds and aquaria or encouraging the decontamination of equipment between usages in different waterbodies. Preventive management of parrot's feather spread by means of legislation or codes of conduct is discussed under 'Reduction of trade through legislation and codes of conduct'.

### 1.5.4.3 Reduction of trade through legislation and codes of conduct

- One randomized, before-and-after trial in the Netherlands<sup>2</sup> reported that the implementation of a code of conduct reduced the trade of aquatic plants banned from sale (group that included parrot's feather *Myriophyllum aquaticum*).
- One study in the USA<sup>1</sup> found that parrot's feather plants were still traded despite a state-wise trade ban.

#### Background

Trade acts as the most important introduction pathway for the entry of invasive aquatic plants (Brunel 2009; Hussner *et al.* 2014). Legislation and voluntary codes of conduct (Verbrugge *et al.* 2014) can help to prevent new introductions and further spread of introduced invasive aquatic plants such as parrot's feather, and thus lessen current and future impacts and associated management costs. Preventive management of parrot's feather by education programmes is discussed under 'Public education'.

Brunel S. (2009) Pathway analysis: aquatic plants imported in 10 EPPO countries. *EPPO Bulletin* 39, 201–213.

Hussner A., Nehring S. & Hilt S., (2014) From first reports to successful control: a plea for improved management of alien aquatic plant species in Germany. *Hydrobiologia* 737, 321–331.

Verbrugge L.N.H., Leuven R.S.E.W., Van Valkenburg J.L.C.H. & van den Born R. (2014) Evaluating stakeholder awareness and involvement in risk prevention of aquatic invasive plant species by a national code of conduct. *Aquatic Invasions*, 9, 369–381.

Hussner A., Stiers I., Verhofstad M.J.J.M., Bakker E.S., Grutters B.M.C., Haury J., van Valkenburg J.L.C.H., Brundu G., Newman J., Clayton J.S. & Anderson L.W.J. (2017) Management and control methods of invasive alien freshwater aquatic plants: A review. *Aquatic Botany*, 136, 112-137.

A study between 2008 and 2010 in Connecticut, USA (1) reported that banning the trade of parrot's feather *Myriophyllum aquaticum* did not eliminate the trade in the species. After a state-wide trade ban, parrot's feather was available for sale in two out of 23 stores surveyed in 2008 (9%) and in one out of 47 stores surveyed in 2010 (2%). Additionally, in 2010, five stores sold a *Myriophyllum* species that could not be identified through morphological or molecular techniques. Nearly 30% of the stores surveyed sold aquatic plants banned in the state of Connecticut. At each store, authors purchased any aquatic plants that morphologically resembled a species banned in Connecticut. The species of the specimens purchased was identified morphologically and through DNA sequencing. Authors did not present the date of the trade ban.

A randomized, before-and-after trial between 2010 and 2012 in the Netherlands (2) reported that the implementation of a code of conduct reduced the trade of aquatic plants banned from sale (group that included parrot's feather *Myriophyllum aquaticum*). The number of batches of banned species found per store visited was higher in 2010 (prior to the implementation of the code of conduct; 0.72 batches/store visited), than in 2011 and 2012 (after the implementation of the code of conduct; 0.03 batches/store visited). Results were not subject to statistical tests. Number of addresses selling aquatic plants visited was 133 in 2010, 107 in 2011 and

76 in 2012. In addition to parrot's feathers, species banned in the Netherland and counted during the study included *Crassula helmsii*, esthwaite waterweed *Hydrilla verticillata*, floating pennywort *Hydrocotyle ranunculoides*, water primrose *Ludwigia grandiflora*, creeping water-primrose *Ludwigia peploides* and variable-leaf watermilfoil *Myriophyllum heterophyllum*. The code of conduct aimed to reduce the introduction and spread of invasive aquatic plants and was developed in partnership between the government and the horticulture sector.

(1) June-Wells M., Vossbrinck C.R., Gibbons J. & Bugbee G. (2012). The aquarium trade: a potential risk for nonnative plant introductions in Connecticut, USA. *Lake and Reservoir Management*, 28, 200-205.

(2) Verbrugge L.N.H., Leuven R.S.E.W., Van Valkenburg J.L.C.H. & van den Born R. (2014) Evaluating stakeholder awareness and involvement in risk prevention of aquatic invasive plant species by a national code of conduct. *Aquatic Invasions*, 9, 369-381.

### **1.5.5 Multiple integrated measures**

- We found no evidence on the use of multiple integrated measures to control parrot's feather.

#### **Background**

Due to the complexity of controlling parrot's feather, multiple integrated measures may be needed for the complete removal of the species from one site. These could include a combination of mechanical and physical control techniques, biological control techniques and/or chemical control techniques. For example, suction dredging and hand-weeding could be used in combination for the removal of submerged invasive aquatic plants, or herbicide could be applied prior to grass carp stocking for faster control and a reduction of management costs (De Winton *et al.* 2013).

De Winton M., Jones H., Edwards T., Özkundakci D., Wells R., McBride C., Rowe D., Hamilton D., Clayton J., Champion P. & Hofstra D. (2013) *Review of Best Management Practices for Aquatic Vegetation Control in Stormwater Ponds, Wetlands, and Lakes*. Auckland Council technical report, TR2013/026.

## 2 Invasive molluscs

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### 2.1 Asian clams

#### Background

Asian clams *Corbicula* spp. can dominate the bed of rivers, displacing native organisms. Their filtering capacity can increase water clarity and lead to an increase in growth of bottom-rooting plants. Occasional mass die-offs of Asian clams can result in deoxygenation of the water that can have knock-on impacts on entire ecosystems (Sousa *et al.* 2008). Live animals and empty shells can cause blockages of channels and pipes serving waterworks, power plants and irrigation systems (Isom 1986).

The taxonomy of Asian clams is complicated by its possession of a rare form of sexual reproduction. In the UK, one phenotype commonly known as *Corbicula fluminea* is present in a number of waterbodies. A second phenotype, known as *Corbicula fluminalis*, is known from mainland Europe but has not yet been recorded from the UK (Pigneur *et al.* 2011).

Isom B.G. (1986) Historical review of Asiatic clam (*Corbicula*) invasion and biofouling of waters and industries in the Americas. *American Malacological Bulletin Special Edition*, 2, 1-5.

Pigneur L.-M., Marescaux J., Roland K., Etoundi E., Descy J.-P. & Van Doninck K. (2011) Phylogeny and androgenesis in the invasive *Corbicula* clams (Bivalvia, Corbiculidae) in Western Europe. *BMC Evolutionary Biology*, 11, 147.

Sousa R., Antunes C. & Guilhermino L. (2008) Ecology of the invasive Asian clam *Corbicula fluminea* (Müller, 1774) in aquatic ecosystems: an overview. *Annales de Limnologie*, 44, 85-94.

#### Key messages

##### ***Drain the invaded waterbody***

No evidence was captured on the effects of draining in the control of Asian clams.

##### ***Exposure to parasites***

No evidence was captured on the effects of exposure to parasites in the control of Asian clams.

##### ***Exposure to disease-causing organisms***

No evidence was captured on the effects of exposure to disease causing organisms in the control of Asian clams.

##### ***Reduce oxygen in water***

A controlled laboratory study from the USA found that Asian clams were not susceptible to low oxygen levels in the water.

##### ***Change pH of water***

No evidence was captured on the effects of pH change in the control of Asian clams.

##### ***Change salinity of water***

A controlled, replicated laboratory study from the USA found that exposure to saline water killed all Asian clams.

### ***Change temperature of water***

A controlled laboratory study from the USA found that exposure to water at temperatures of 37°C and 36°C killed all Asian clams within 2 and 4 days, respectively.

### ***Use of gas-impermeable barriers***

One controlled study from North America found that placing gas impermeable fabric barriers on a lake bottom (several small and one large area) reduced populations of Asian clams.

### ***Add chemicals to the water***

Two replicated laboratory studies and one controlled, replicated field study found that chlorine, potassium and copper killed Asian clams. Increasing chemical concentration and water temperature killed more clams in less time. One controlled field trial achieved 80% and 100% mortality of Asian clams using encapsulated control agents (SB1000 and SB2000 respectively) in irrigation systems.

### ***Cleaning equipment***

A field study from Portugal found that mechanical removal, followed by regular cleaning and maintenance of industrial pipes at a power plant permanently removed an Asian clam population. A field study from Portugal found that adding a sand filter to a water treatment plant reduced an Asian clam population.

### ***Mechanical removal***

A controlled before-and-after study from North America found suction dredging of sediment reduced an Asian clam population by 96%, and these effects persisted for a year. A replicated, controlled, before-and-after field trial in Ireland showed that three types of dredges were effective at removing between 74% and >95% of the Asian clam biomass.

### ***Hand removal***

No evidence was captured on the effects of hand removal in the control of Asian clams.

### ***Public awareness and education***

No evidence was captured on the effects of raising public awareness or education in the control Asian clams.

## **2.1.1 Drain the invaded water body**

- No evidence was captured for the use of dewatering as a management tool for Asian clams.

### **Background**

The relatively thick shells of Asian clams *Corbicula* spp. will offer protection against desiccation, but prolonged periods of dewatering may serve as an effective management tool in some situations. Large die-offs of Asian clams have been reported during droughts (Ilarri *et al.* 2011). Also, an ecological study conducted between 2005 and 2006 in Lake Constance, which borders Germany, Switzerland and

Austria (Werner & Rothhaupt 2008) found that an Asian clam *Corbicula fluminea* population was adversely affected by unprecedented low water levels and a harsh winter with consequently low water temperatures. Low water levels led to 100% mortality in clams situated at mean low-water level due to desiccation. At lower depths, water temperatures as low as 2°C over a three month period led to a mass die out of the clams. Juvenile clams survived longer than adults. However, at the end of winter, only 1% of the population remained. Although an intervention was not directly tested in this study, the results suggest that low temperatures and drainage may be possible methods of controlling Asian clam populations in the wild. In regulated rivers, it may also be possible to decrease the flow rate in order to reduce the water level and control Asian clam populations.

Bódis E., Tóth B. & Sousa R. (2014) Massive mortality of invasive bivalves as a potential resource subsidy for the adjacent terrestrial food web. *Hydrobiologia*, 735, 253-262.

Ilarri M., Antunes C., Guilhermino L. & Sousa R. (2011) Massive mortality of the Asian clam *Corbicula fluminea* in a highly invaded area. *Biological Invasions*, 13, 277-280.

Werner S. & Rothhaupt K.-O. (2008) Mass mortality of the invasive bivalve *Corbicula fluminea* induced by a severe low-water event and associated low water temperatures. *Hydrobiologia*, 613, 143-150.

### **2.1.2 Exposure to parasites**

- No evidence was captured for the use of parasite exposure to control Asian clams.

#### **Background**

Parasites have the potential to control clam populations by reducing production of offspring, increasing risk of predation, slowing growth rates or causing death. Studies in Asian clams *Corbicula* spp. have focussed on their use as a tool for monitoring water-borne parasites of interest to humans, rather than identifying parasites that may regulate clam populations.

### **2.1.3 Exposure to disease-causing organisms**

- No evidence was captured for the use of exposure to disease-causing organisms for the control of Asian clams.

#### **Background**

Disease-causing organisms have the potential to control Asian clam *Corbicula* spp. populations by reducing reproductive outputs, increasing risk of predation, slowing growth rates or causing death. Studies in Asian clams have focussed on their use as a

tool for monitoring water-borne pathogens of interest to humans, rather than identifying pathogens that may regulate clam populations.

#### **2.1.4 Reduce oxygen in the water**

- A controlled laboratory study conducted in the USA<sup>1</sup> found that Asian clams were resistant to extreme very low levels of oxygen, irrespective of water temperature or length of immersion in the test conditions.

##### **Background**

Very low levels of oxygen in the water for a prolonged period may kill Asian clams *Corbicula* spp. However, even mild reductions in oxygen may offer advantages by increasing the vulnerability of the clams to predation. When attacked, bivalves protect their soft tissues by closing their protective valves. This reduces vulnerability to small predators, but ventilation and oxygen uptake are suspended. In a laboratory study it was found that after a simulated attack, Asian clams under low oxygen conditions reopened their valves sooner than clams under high oxygen conditions, suggesting that low oxygen levels increases vulnerability to predation (Saloom & Duncan 2005).

Saloom M.E. & Duncan R.S. (2005) Low dissolved oxygen levels reduce anti-predation behaviours of the freshwater clam *Corbicula fluminea*. *Freshwater Biology*, 50, 1233-1238.

A controlled laboratory study in 1999 on specimens from a dam and artificial stream in Texas, USA<sup>1</sup> found that Asian clams *Corbicula fluminea* survived low oxygen levels for extended time periods. They survived an average of 12, 35 and >84 days at 25°C, 15°C and 5°C, respectively. Survival rates were comparable with the control (normal oxygen levels). However, larger clams were less tolerant to low oxygen than smaller ones. Groups of clams were acclimated to 5°, 15° or 25°C for 14 days. A group of 30 adult clams were held in water that was either aerated (control) or had reduced oxygen at 5°, 15° and 25°C. In low oxygen treatments, partial pressure of oxygen was reduced to less than 5% of full air saturation by continually bubbling the water with nitrogen. The water was changed every 2-3 days. Testing ceased when all clams had died or after a maximum of 12 weeks. Oxygen concentrations and survival of clams were recorded daily.

(1)Matthews M.A. & McMahon R.F. (1999) Effects of temperature and temperature acclimation on survival of zebra mussels (*Dreissena polymorpha*) and Asian clams (*Corbicula fluminea*) under extreme hypoxia. *Journal of Molluscan Studies*, 65, 317-325.

### **2.1.5 Change pH of the water**

- No evidence was captured for the use of pH adjustment to control Asian clams.

#### **Background**

The ability of Asian clams *Corbicula* spp. to lay down their calcium-rich shells will be reduced in more acidic (lower pH) conditions. Molluscs are rarely found in waters with a pH of less than 6.0-6.5 and the use of acid dosing to lower pH has been used to effectively control other invasive bivalves (Claudi *et al.* 2012).

Claudi R., Graves A., Taraborelli A.C., Prescott R.J. & Mastitsky E. (2012) Impact of pH on survival and settlement of dreissenid mussels. *Aquatic Invasions*, 7, 21-28.

### **2.1.6 Change salinity of the water**

- A controlled, replicated laboratory study conducted in the USA<sup>1</sup> found that Asian clams were killed (100% mortality) when exposed to high salinities (18-34‰).

#### **Background**

Elevated levels of salt in the water can cause osmotic stress in Asian clams *Corbicula* spp., leading to mortality. Increasing the salinity of a waterbody to a very high level is likely to have significant effects on non-target species.

A controlled, replicated laboratory study conducted in 1978 on specimens from a stream in Florida, USA<sup>1</sup> found that placing Asian clams *Corbicula* spp. in high salinity water resulted in 100% mortality. Clams survived in salinities of up to 10‰ for two months. Salinities between 18 and 34‰ led to 100% mortality within 10 days. Clams were transferred from a stream to 10 x 6 litre tanks containing stream water to acclimate. Ten clams were placed in each treatment tank. Following initial acclimation, the clams were placed in salinities from 0.5-40 parts per thousand. Valve movements were recorded and survival checked daily. Salinity levels were considered lethal if mortality was observed in at least 50% of the clams.

(1)Gainey L.F. (1978) The Response of the Corbiculidae (Mollusca: Bivalvia) to Osmotic Stress: The Organismal Response. *Physiological Zoology*, 51, 68-78.

### **2.1.7 Change temperature of the water**

- A controlled laboratory study in the USA<sup>1</sup> found that temperatures of 36°C or higher killed Asian clams within or after four days.

## Background

All organisms will have an upper thermal tolerance and so raising water temperatures may offer a tool for management of Asian clams *Corbicula* spp. Such an approach may be especially suited to industrial facilities that generate an excess of heated water, such as power plants. Reducing water temperatures can also offer a possible management tool. Studies in the St. Clair River, USA reported very large winter die-offs of *Corbicula fluminea* (French & Schloesser 1996). Populations persisted in the waters immediately downstream of a power plant in which water temperature was relatively high.

French J.R.P. & Schloesser D.W. (1996) Distribution and winter survival health of Asian clams, *Corbicula fluminea*, in the St. Clair River, Michigan. *Journal of Freshwater Ecology*, 11, 183-192.

A controlled laboratory study conducted between 1976 and 1977 on specimens from a river in Virginia, USA<sup>1</sup> found that exposure to temperatures of 36°C and higher killed Asian clams *Corbicula fluminea*. All clams were dead after either four days at 36°C or two days at 37°C compared with clams surviving in a control treatment at 25°C. Clams were placed in seven heated aquatic chambers, plus one control chamber. In total, 19 clams were placed in each chamber. Over a 24-hour period, infrared lamps raised chamber temperatures to 27, 29, 31, 33, 35, 36 and 37°C. These temperatures were held for four days. A control group was maintained at 25°C. Mortality status of clams was checked and recorded.

(1) Cherry D.S., Rodgers J.H., Graney R.L. & Cairns J. (1980) Dynamics and control of the Asiatic clam in the New River, Virginia. *Bulletin of the Virginia Water Resources Center*, 123, 1-72.

### 2.1.8 Use gas-impermeable barriers

- A controlled study in North America<sup>1</sup> found that placing gas-impermeable barriers across the bottom of the lake (several small fabric covers or one large cover) significantly reduced the abundance of Asian clams.

## Background

Gas-impermeable barriers can be placed on the river or lake bed as a possible management tool for Asian clams *Corbicula* spp.. As organisms beneath die, oxygen levels fall and this can result in clam death.

A controlled, replicated study conducted in 2009-2011 in Lake Tahoe, North America<sup>1</sup> found that gas-impermeable fabric laid across the lake bottom killed 98-100% of Asian clams *Corbicula fluminea* after 30-120 days. During one trial all clams were killed after one month, compared to only 3% in control plots without fabric. During the second trial the abundance of Asian clams after 120 days under gas-

impermeable fabric was 98% lower than clam abundance in control plots. One year after the barrier was removed from this second trial, clam abundance was still 90% lower compared to control plots. Mortality was caused by a reduction in dissolved oxygen concentrations, which dropped to zero after 72 hours during the first trial and after eight days in the second trial. In 2009, six plastic (ethylene propylene diene monomer) barriers (9m<sup>2</sup>; 1 mm thick) were placed by scuba divers on the lake bottom at 5 m water depth and left in place for 4-56 days. In 2010-2011 a plastic barrier (1,950m<sup>2</sup>; 1 mm thick) was placed by scuba divers on the lake bottom at 5m depth and left for 120 days. The survival of clams underneath the fabric test sites and in control plots was monitored.

(1) Wittmann M.E., Chandra S., Reuter J.E., Schladow S.G., Allen B.C. & Webb K.J. (2012) The control of an invasive bivalve, *Corbicula fluminea*, using gas impermeable benthic barriers in a large natural lake. *Environmental Management*, 49, 1163-1173.

### **2.1.9 Add chemicals to the water**

- A replicated laboratory study in the USA<sup>1</sup> found that dosing with the biocides chlorine, potassium and copper killed Asian clams.
- A controlled, replicated laboratory study and a controlled, replicated field study in the USA<sup>2</sup> found that higher concentrations of chlorine and bromine, delivered at higher temperatures, shortened the time required to kill the Asian clams.
- A controlled field-based trial in Spanish irrigation systems<sup>3</sup> showed that fat-coated particles called BioBullets could kill 100% of the Asian clams within pipes.

#### **Background**

Many chemicals will be effective at killing Asian clams *Corbicula* spp., but the wider impacts on non-target organisms must also be taken into account. Control agents that do not persist or accumulate in the wider environment are preferable, as are agents that might offer some specificity to bivalve molluscs. Combinations of control agents can offer particular benefits if they enhance one another's toxicity effect upon the clams.

A replicated laboratory study conducted between 1976 and 1978 on specimens from a river in the USA<sup>1</sup> found that Asian clams *Corbicula fluminea*, were killed when exposed to concentrations of chlorine, potassium and heavy metals. Half of the clams died after exposure to 0.69 mg per litre of chlorine in tanks for 10 days. Up to 89% died within four days of exposure to 140 mg per litre of potassium. Copper was the most toxic with half of the clams dying when exposed to 0.59 mg per litre for one day and 0.04 mg per litre for four days. The toxicity of several biocides was tested in static and continuous-flow tests. Fourteen replicate tanks and four artificial streams were used, respectively. There were 12 clams per tank and nine clams per artificial stream (flow rates were maintained at 0.5-0.8 litres/min). Clams were exposed to

various concentrations of chlorine, potassium and copper for between one and 10 days. Survival of clams was checked and recorded.

A controlled, replicated laboratory and field study in 1983-1985 in the USA<sup>2</sup> found that dosing clams with chlorine and bromine killed up to 95% of Asian clams *Corbicula fluminea*. Dosing at higher concentrations and higher temperatures killed clams in a shorter time. Adults and juveniles were similarly sensitive to both chlorine and bromine and both chemicals were equally effective. In the laboratory, < 53% of clams died when exposed to a 32 day dose at 0.2-1.0 mg/litre total residual chlorine (TRC) at 16 °C. Using the same doses and duration but at higher temperatures (>18 °C) killed > 53% of clams. In the laboratory, a 14 day low dose (0.25 mg/litre TRC) followed by an 18 day high dose (0.5-1.0 mg/litre TRC) killed > 80% of the clams at 20 °C. A constant high dose (0.5-1.0 mg/litre TRC) for 32 days at 20 °C killed 60-95% of the clams. In the field, 90% of clams were killed when exposed to a 28 day dose of 0.25 mg/litre TRC during the spring when ambient temperatures were 20-25 °C. In the autumn, <24% of clams were killed when exposed to a 28 day dose of <0.5 mg/litre TRC, when ambient temperatures were lower (12-20 °C). No clams died in the control treatments. In the laboratory tests, 30 adult clams were placed in one of five replicate artificial streams with chlorine treatments. Juveniles were added to some replicates. Survival of clams was recorded daily. In the field, two flow-through chambers (1 x 0.25 m) in spring and three in autumn were used to expose 25-30 Asian clams with chlorinated water. Chambers were placed at four sites within the intake stream of an industrial plant which suffered from Asian clam fouling. A control group of clams was exposed to non-chlorinated water. Survival of clams was recorded.

A controlled field trial conducted in 2011 in irrigation systems in Spain<sup>3</sup> found that Asian clams of *Corbicula fluminea* and *Corbicula fluminalis* species showed 80% mortality when exposed to a fat-coated chemical called SB1000 and 100% mortality when exposed to a fat-coated chemical called SB2000. Clams of all sizes present were equally susceptible to the chemical. The chemicals were coated with an attractant to the clams. This targeted method of delivery to the filter-feeding clams resulted in the need for reduced concentrations of the chemicals. The product was approved for use within in-service irrigation systems for almonds, cherries and olives, and there were no negative effects on the crops. The chemical SB1000 was dosed at 150 mg/l and SB2000 at 30 mg/l for eight hours a day for two days, using a calibrated powder doser, with the products being delivered into different parts of the irrigation system. The number of live, freshly gaping and freshly empty shells were monitored before and after dosing and subsamples of clams were measured. Clams were monitored in a control irrigation pipe which received no chemicals. No clams died in the control pipe.

(1) Cherry D.S., Rodgers J.H., Graney R.L. & Cairns J. (1980) Dynamics and control of the Asiatic clam in the New River, Virginia. *Bulletin of the Virginia Water Resources Center*, 123, 1-72.

(2) Doherty F.G., Farris J.L., Cherry D.S. & Cairns J. (1986). Control of the freshwater fouling bivalve *Corbicula fluminea* by halogenation. *Archives of Environmental Contamination and Toxicology*, 15, 535-542.

(3) BioBullets (2012) *BioBullets para el control de obstrucciones de mejillon cebra en el systems de regadio Espanol*. Report for Confederación Hidrográfica del Ebro 1-14

### **2.1.10 Clean equipment**

- A study in Portugal<sup>1</sup> found that mechanical removal and regular cleaning of industrial pipes or addition of a sand filter were effective methods of permanently removing or reducing numbers of Asian clams, respectively.

#### **Background**

The economic impacts of Asian clams *Corbicula* spp. might be reduced by the regular cleaning of vulnerable equipment and machinery. Cleaning can often be easily managed and cost-effective.

A study conducted in 1980-2010 at a power station and drinking water treatment plant in Portugal<sup>1</sup> found that subjecting structures infested by the Asian clam *Corbicula fluminea* to cleaning, maintenance and sand filtration prevented or reduced re-infestation. At the power station, cleaning and maintenance procedures completely removed clam populations from the bypass channel of the power station and prevented re-infestation. At the treatment plant, installation of a multilayer sand-filter downstream from the raw water reservoir significantly reduced the amount of Asian clams passing through into the waterworks. Of the facilities managers interviewed, three out of 420 drinking water plants and two out of six power plants provided details on clam control. Managers were interviewed about past or current occurrence of clam infestation episodes, the types of structures affected, interventions in place and their degree of success. Interventions included mechanically removing and washing out clams, shortening the period between filter maintenance and regularly replacing sand in the multilayer sand filter.

(1) Rosa I.C., Pereira J.L., Gomes J., Saraiva P.M., Goncalves F. & Costa R. (2011) The Asian clam *Corbicula fluminea* in the European freshwater-dependent industry: A latent threat or a friendly enemy? *Ecological Economics*, 70, 1805-1813.

### **2.1.11 Mechanical removal**

- A replicated, controlled, before-and after trial in North America<sup>1</sup> found that suction dredging reduced Asian clam densities within the sediment by 96% over two weeks and that the reduction persisted for a year.
- A replicated, controlled, before-and-after field trial in Ireland<sup>2</sup> found that three types of dredges were equally effective at removing Asian clams, resulting in a biomass reduction ranging from 74% to >95%, and an density reduction ranging from 65% to 95%.

## Background

Technology for collecting clams from sediments has been well-developed in commercial shellfisheries. Such tools can be employed within open water systems to remove Asian clams *Corbicula* spp.

A replicated, controlled, before-and-after study conducted between 2009 and 2010 at two lake sites in North America<sup>1</sup> found that suction dredging significantly reduced the abundance of the Asian clam *Corbicula fluminea* compared to control (non-dredged) sites. After two weeks, density was reduced from around 1,500 to 60 clams/m<sup>2</sup> (96% reduction). These effects lasted for at least a year. Diver-assisted suction dredging was applied in five metres water depth at two sites. The equipment had a 4 cm diameter hose, 5.5 Horse Power engine at 3,600 rpm net power output, and 196 cm<sup>3</sup> displacement. Each site had three dredged (to 8-13 cm deep) and one un-dredged control plot of 36 m<sup>2</sup>.

A replicated, controlled, before-and-after field trial conducted during 2012 in the tidal reaches of the River Barrow, Ireland<sup>2</sup> found that dredging could reduce the biomass and density of the Asian clam *Corbicula fluminea*. At a site with high clam biomass and high clam density, dredging achieved a reduction of greater than 95% biomass and 95% clam density. At a site with a low density and low biomass of clams dredging achieved a reduction of biomass by 82% and density by 65%. At a site with high density and low biomass of clams dredging achieved a reduction of biomass by 74% and density by 92%. There was no difference in the effectiveness of the three dredge types used. In each of the three sites, three control and three experimental plots were marked by buoys. In each plot, clam biomass and density was estimated before and after trials using five 0.25m<sup>2</sup> quadrats which were hand-searched by divers. The three experimental plots at each site were dredged using either a box dredge, an electric dredge or a hydraulic dredge. Dredging selectively removed larger clams (18-32 mm length).

(1) Wittmann M.E., Chandra S., Reuter J.E., Caires A., Schladow S.G. & Denton M. (2012) Harvesting an invasive bivalve in a large natural lake: Species recovery and impacts on native benthic macroinvertebrate community structure in Lake Tahoe, USA. *Aquatic Conservation: Marine and Freshwater Ecosystems*, 22, 588-597.

(2) Sheehan R., Caffrey J.M., Millane M., McLoone P., Moran H. & Lucy, F. (2014) An investigation into the effectiveness of mechanical dredging to remove *Corbicula fluminea* (Müller, 1774) from test plots in an Irish river system. *Management of Biological Invasions*, 5, 407-418.

### 2.1.12 Remove by hand

- No evidence was captured on the use of hand removal as a tool for managing Asian clams.

## Background

In shallow waters it may be possible to collect Asian clams *Corbicula* spp. by hand, and especially large individuals. Disturbance of the sediment results in the animals becoming visible at the surface.

### 2.1.13 Public awareness and education

- No evidence was captured on the use of raising public awareness and education as tools for managing Asian clams.

## Background

In order to minimize the transport of clams by humans, measures should be taken such as the education of fishermen in not using Asian clams *Corbicula* spp. as bait outside invaded places (Aldridge & Müller 2001) and the possibility of small specimens hitchhiking in their clothes and boats. Caution should be taken in not transferring sand or gravel from invaded locations (Counts 1986).

Aldridge D.C. & Müller S.J. (2001) The Asiatic clam, *Corbicula fluminea*, in Britain: current status and potential threats. *Journal of Conchology*, 37, 177-184.

Counts C.L. III (1986) The zoogeography and history of the invasion of the United States by *Corbicula fluminea* (Bivalvia: Corbiculidae). *American Malacological Bulletin, Special Edition No. 2*, 7-39.

## 3 Invasive crustaceans

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### 3.1 Ponto-Caspian gammarids

#### Background

The Ponto-Caspian gammarids of concern belong to the genera *Echinogammarus*, *Dikerogammarus*, *Pontogammarus* and *Obesogammarus*. Western Europe has suffered a mass invasion of this taxon in recent years. This is a result of canal constructions that have linked the Caspian and Black Seas to the Rhine and Danube watersheds. Ponto-Caspian gammarids are predicted to establish widely across the UK (Gallardo & Aldridge 2013). They can attain high densities and wide distributions. Ponto-Caspian gammarids predate upon native gammarids, fish eggs and macroinvertebrates. They can also switch their feeding to focus on algae and leaf material. Because of this, they drive changes in the community structure of invaded systems and can also affect the distribution of fishes that may feed upon them. Several interventions have been tested within freshwater habitats, to control or eradicate invasive Ponto-Caspian gammarids from invaded systems.

Gallardo, B. & Aldridge, D.C. (2013) Priority setting for invasive species management: risk assessment of Ponto-Caspian invasive species into Great Britain. *Ecological Applications*, 23, 352-364.

#### Key messages

##### ***Biological control using predatory fish***

No evidence was captured on the use of predatory fish as a biocontrol agent to control Ponto-Caspian gammarids.

##### ***Control movement of gammarids***

Two replicated studies, including one controlled study, in the USA and UK found that movements of invasive freshwater shrimp slowed down or were stopped when shrimp were placed in water that had been exposed to predatory fish or was carbonated.

##### ***Exposure to parasites***

A replicated, controlled experimental study in Canada found that a parasitic mould reduced populations of freshwater invasive shrimp.

##### ***Exposure to disease-causing organisms***

No evidence was captured on the use of disease-causing organisms to control Ponto-Caspian gammarids.

##### ***Change salinity of water***

One of two replicated studies, including one controlled study, in Canada and the UK found that increasing the salinity level of water killed the majority of invasive shrimp within five hours. One found that increased salinity did not kill invasive killer shrimp.

### **Change water temperature**

A controlled laboratory study from the UK found that heating water in excess of 40°C killed invasive killer shrimps.

### **Change water pH**

A controlled laboratory study from the UK found that lowering the pH of water did not kill invasive killer shrimp.

### **Dewatering (drying out) habitat**

A replicated, controlled laboratory study from Poland found that lowering water levels in sand (dewatering) killed three species of invasive freshwater shrimp, although one species required water content levels of 4% and below before it was killed.

### **Add chemicals to water**

A controlled laboratory study from the UK found that four of nine substances added to freshwater killed invasive killer shrimp, but were impractical (iodine solution, acetic acid, Virkon S and sodium hypochlorite). Five substances did not kill invasive shrimp (methanol, citric acid, urea, hydrogen peroxide and sucrose).

### **Cleaning equipment**

No evidence was captured on the use equipment cleaning to control Ponto-Caspian gammarids.

### **Exchange ballast water**

No evidence was captured on the use of ballast water exchange to control Ponto-Caspian gammarids.

## **3.1.1 Biological control using predatory fish**

- No evidence was captured for the use of predatory fish to control Ponto-Caspian gammarids.

### **Background**

Predatory fish could potentially be used to biologically control invasive shrimp. A study from Lake Constance, which borders Germany, Switzerland and Austria, found that the arrival of the invasive Ponto-Caspian killer shrimp *Dikerogammarus villosus* led to several fish species in the lake changing their diet (Eckmann et al. 2008). Burbot *Lota lota*, European eel *Anguilla anguilla* and Eurasian perch *Perca fluviatilis* all fed on the killer shrimp (Eckmann et al. 2008).

Eckmann R., Mortl M., Baumgartner D., Berron C., Fischer P., Schleuter D. & Weber A. (2008) Consumption of amphipods by littoral fish after the replacement of native *Gammarus roeseli* by invasive *Dikerogammarus villosus* in Lake Constance. *Aquatic Invasions*, 3, 187-191.

### 3.1.2 Control movement of gammarids

- A replicated, controlled laboratory study in the USA<sup>1</sup> found that movements of invasive freshwater shrimp slowed down or stopped when they were placed in water that had been exposed to different species of predatory fish, compared to those not exposed to fish.
- A replicated laboratory study in the UK<sup>2</sup> found carbonating the water stunned invasive killer shrimp.

#### Background

Containment of invasive gammarids can help to prevent their spread both within the invaded waterway and also reduce the likelihood of them being transported into new waterbodies.

A replicated, controlled experimental study conducted in 2008, on specimens from a canal in the USA<sup>1</sup> found that movement of invasive freshwater shrimp *Echinogammarus ischnus*, slowed down or stopped when exposed to water that had previously contained predatory fishes. Movement patterns were significantly lower when compared with a control treatment, i.e. fish-free water. Increased avoidance behaviour was associated with increased density of fishes previously in the water. The treatment water had been in contact with either the round goby *Apollonia melanostoma*, yellow perch *Perca flavescens*, black crappie *Promoxis nigromaculatus*, rainbow darter *Etheostoma caeruleum* or the brown bullhead *Ameiurus nebulosus*. One shrimp was put in each of two tanks (7 x 7 cm) containing 1 cm deep freshwater. One tank was fed with water from a 40 l tank containing 1-10 fishes at 2 ml/second over a 30 second period. The other tank was fed with water without fish. Shrimp movements were observed and measured.

A controlled laboratory study conducted in 2011 in England, UK<sup>2</sup> found that carbonating water did not kill the invasive killer shrimp *Dikerogammarus villosus* but 100% of the shrimps were stunned (stopped moving). A test group of five captive shrimp was immersed in carbonated water for 15 minutes. Dead and live shrimp were counted.

(1) Pennuto C. & Keppler D. (2008) Short-term predator avoidance behaviour by invasive and native amphipods in the Great Lakes. *Aquatic Ecology*, 42, 629-641.

(2) Stebbing P.D., Sebire M. & Lyons B. (2011) Evaluation of a number of treatments to be used as biosecurity measures in controlling the spread of the invasive killer shrimp (*Dikerogammarus villosus*). CEFAS Final Contract Report C5256. 40 pp.

### 3.1.3 Exposure to parasites

- A replicated, laboratory study in Canada<sup>1</sup> found that an introduced parasitic mould reduced populations of an invasive shrimp.

## Background

Parasites have the potential to control gammarid populations by reducing production of offspring, increasing risk of predation, slowing growth rates or causing death. In the wild, parasite infections have been associated with rapid die-offs of natural populations of freshwater shrimps, with the invasive species being more vulnerable than native species (Kestrup *et al.* 2011).

Kestrup A.M., Thomas S.H., van Rensburg K., Ricciardi A. & Duffy M.A. (2011) Differential infection of exotic and native freshwater amphipods by a parasitic water mould in the St. Lawrence River. *Biological Invasions*, 13, 769-779.

A replicated, controlled laboratory study in 2011 in Canada<sup>1</sup> found that a parasitic water mould (oomycete) of unknown origin infected and killed invasive gammarids *Echinogammarus ischnus*. Invasive shrimps exposed to water carrying the mould had a 52% mortality rate after seven days, compared with 16% mortality in native shrimps. Laboratory tests used 20 replicate aquaria each containing 10 invasive and 10 native (*Gammarus fasciatus*) shrimps. Two litres of river water was placed in each aquarium from the St. Lawrence River, which was the location of the mould's original discovery. Aquaria were checked twice daily for seven days and dead individuals were removed.

(1)Kestrup A.M., Thomas S.H., van Rensburg K., Ricciardi A. & Duffy M.A. (2011) Differential infection of exotic and native freshwater amphipods by a parasitic water mould in the St. Lawrence River. *Biological Invasions*, 13, 769-779.

### 3.1.4 Exposure to disease-causing organisms

- No evidence was captured for the use of disease-causing organisms to control Ponto-Caspian gammarids.

## Background

Disease-causing organisms (pathogens) have the potential to control gammarid populations by reducing reproductive outputs, increasing risk of predation, slowing growth rates or causing death.

### 3.1.5 Change salinity of the water

- One of two replicated laboratory studies (one controlled) in Canada<sup>1</sup> and the UK<sup>2</sup> found that increasing the salinity level of water killed the majority of invasive shrimp within five hours. One found that increased salinity did not kill invasive killer shrimp<sup>2</sup>.

## Background

Freshwater gammarid shrimps are sensitive to increased levels of salt in the water. Addition of salt to the water, or exposure to sea water therefore offers a potential management tool.

A replicated, controlled laboratory experiment in 2009, on specimens taken from a river in Canada<sup>1</sup> found that invasive freshwater shrimp *Echinogammarus ischnus*, were killed when salt was added to water. Within five hours, 66% of shrimp died from exposure to saline water (30% salinity). It did not make a difference if the water salinity was increased gradually or immediately. Only 33% of shrimp treated survived for up to two days and 0% beyond two days. Ten shrimp were placed in each of 12 glass jars. The water in four of the jars had 30% salinity from the beginning (using unfiltered river water). The salinity in another four jars was 4% at the start of the experiment and increased every hour to 8, 14, 24 and 30%. Four jars were controls (freshwater only). Every hour for five hours, and after 24 and 48 hours, dead animals were removed and live animals counted.

A controlled laboratory study conducted in 2011 in England, UK<sup>2</sup> found that adding salt to freshwater did not kill the killer shrimp *Dikerogammarus villosus*. None of the shrimp died during the test period. This included shrimp exposed to salinity levels 3.5 times more saline than normal seawater. Tests were conducted on 5 captive shrimp immersed for 15 minutes in de-chlorinated water of different salinities ranging from 5 to 160 grams of salt/litre. Artificial marine salt was used to adjust the salinity in the test solutions. Dead and live shrimp were counted.

(1)Ellis S. & McIsaac H.J. (2009) Salinity tolerance of Great Lakes invaders. *Freshwater Biology*, 54, 77-89.

(2)Stebbing P.D., Sebire M. & Lyons B. (2011) Evaluation of a number of treatments to be used as biosecurity measures in controlling the spread of the invasive killer shrimp (*Dikerogammarus villosus*). CEFAS Final Contract Report C5256. 40 pp.

### 3.1.6 Change water temperature

- A controlled laboratory study from the UK<sup>1</sup> found that heating water to >36°C killed all shrimps after 15 minutes exposure and at >43°C all shrimps died immediately.

## Background

The use of warm water in which to dip nets, waders and wetsuits could provide a useful biosecurity tool, and help to reduce the risk of moving gammarids between waterbodies.

A controlled laboratory study in 2011, England, UK<sup>1</sup> found that heating water killed the invasive killer shrimp *Dikerogammarus villosus*. All shrimp died within a 15 minute exposure period when immersed into water at temperatures >36°C. All shrimps died immediately when immersed into water at temperatures >43°C. For each temperature tested, a container holding five shrimp was filled with water with measured temperatures of 28.4, 31.1, 36.3, 38.9, 43.2°C. A control set of five shrimps was held at ambient temperature (14-15°C). Shrimps were observed over a 15 minute period and then moved to freshwater at ambient conditions (14-15°C) to check for mortality over an 80 minute period.

(1) Stebbing P.D., Sebire M. & Lyons B. (2011) Evaluation of a number of treatments to be used as biosecurity measures in controlling the spread of the invasive killer shrimp (*Dikerogammarus villosus*). CEFAS Final Contract Report C5256. 40 pp.

### **3.1.7 Change water pH**

- A controlled laboratory study from the UK<sup>1</sup> found that lowering the pH of water did not kill invasive killer shrimp.

#### **Background**

Making the water more acidic (lowering the pH) through the use of chemicals may kill gammarid shrimps because their exoskeletons are likely to be dissolved by the acid. Prolonged exposure to acidic water may affect the ability of the shrimps to lay down new exoskeletons and to moult.

A controlled laboratory study in 2011 in England, UK<sup>1</sup> found that lowering the pH of water did not kill the killer shrimp *Dikerogammarus villosus*. None of the shrimp died during the test period. Tests were conducted on 5 captive shrimp immersed for 15 minutes in de-chlorinated water of different pH values. Values ranged from pH 7.2-3.1. Hydrochloric acid was used to adjust the pH of the test solutions. Dead and live shrimp were counted.

(1) Stebbing P.D., Sebire M. & Lyons B. (2011) Evaluation of a number of treatments to be used as biosecurity measures in controlling the spread of the invasive killer shrimp (*Dikerogammarus villosus*). CEFAS Final Contract Report C5256. 40 pp.

### **3.1.8 Dewater (dry out) the habitat**

- A replicated, controlled laboratory study from Poland<sup>1</sup> found that lowering water levels in sand killed three species of invasive freshwater shrimp, although one species required water content levels of 4% and below before it was killed.

## Background

Draining invaded waterbodies may offer a tool for localised eradication or population reduction of gammarid shrimps.

A replicated, controlled laboratory study in 2011, on specimens from a reservoir in Poland<sup>1</sup> found that 50-90% of three invasive freshwater shrimp species *Pontogammarus robustoides*, *Dikerogammarus haemobaphes* and *D. villosus* were killed by drying out the sand, with differences in the kill rate explained by the species and level of drying. Only half of the experimental demon shrimps *Dikerogammarus haemobaphes* survived when the water content in the sand was reduced to 9% and only a tenth survived in sand containing 7% water. For the killer shrimp *Dikerogammarus villosus*, only half the shrimp survived in sand containing 11% water and only a tenth survived in sand containing 10% water. The shrimp *Pontogammarus robustoides* was more resistant to drying out. However, once the water content of the sand was below 4%, half of the *P. robustoides* shrimp had died. Five shrimp of each species were put in each of five ceramic trays with a 2 cm sand layer and 1.5 cm water depth. In control trays, water levels were held constant. Treatment trays were left to dry naturally. Dead and alive shrimp were counted daily and water content of the sand was measured by weighing 14 g of the sand before and after drying at 100°C.

(1)Poznańska M., Kakareko T., Krzyżyński M. & Kobak J. (2013) Effect of substratum drying on the survival and migrations of Ponto-Caspian and native gammarids (Crustacea: Amphipoda). *Hydrobiologia*, 700, 47-59.

### 3.1.9 Add chemicals to the water

- A controlled laboratory study in the UK<sup>1</sup> found that iodine solution, acetic acid, Virkon S and sodium hypochlorite added to freshwater killed invasive killer shrimp, but were considered impractical for field application. Methanol, citric acid, urea, hydrogen peroxide and sucrose did not kill invasive killer shrimp when added to freshwater<sup>1</sup>.

## Background

Adding toxic chemical to the water offers the potential for localised control of invasive gammarid shrimps. Chemical 'dips' for fishing and sampling gear can also be an effective biosecurity method for preventing the transport of gammarids between water bodies. Most chemicals will have an impact on non-target organisms, and so their use must be carefully considered before being dosed. In many instances, the use of chemicals may be subject to local or national approval by regulatory authorities.

A controlled laboratory study in 2011 in England, UK<sup>1</sup>, found that when added to freshwater, iodine solution, acetic acid, Virkon S and sodium hypochlorite killed the killer shrimp *Dikerogammarus villosus*, but methanol, citric acid, urea, hydrogen peroxide and sucrose did not. For iodine solution (FAM30) there was 100% mortality within 15 minutes when the shrimp were placed in solutions of 4-6 ml per litre. However, FAM30 is an irritant and so was not considered a practical control method. For acetic acid, a 10% solution was required to kill all the shrimp in 15 minutes. At lower concentrations, no shrimps died during the test period. However, fifteen minutes was considered too long for acetic acid to be a practical control method. For Virkon S, all shrimp exposed to a 1% solution died within 15 minutes, with half dying within eight minutes. However, Virkon S has a relatively short shelf life and a capacity to bleach and can damage equipment and so was not considered a practical control method. For sodium hypochlorite, at 50,000 parts/million, half of the shrimp were killed within 4.5 minutes. However, at that concentration it is lethal to humans and so is not practical. Equipment containing shrimp could be soaked in sodium hypochlorite at 200 parts/million for over an hour, but is considered impractical. None of the shrimps died when exposed for 15 minutes to methanol (1 or 10 %), urea (1 or 10 g/litre), citric acid (15 or 150 mg/litre), hydrogen peroxide (100 mg/litre), or sucrose (10 or 100 g/litre). All tests were conducted on 5 captive shrimp. Dead and live shrimp were counted.

(1)Stebbing P.D., Sebire M. & Lyons B. (2011) Evaluation of a number of treatments to be used as biosecurity measures in controlling the spread of the invasive killer shrimp (*Dikerogammarus villosus*). CEFAS Final Contract Report C5256. 40 pp.

### **3.1.10 Cleaning equipment**

- No evidence was captured for the cleaning of equipment to control Ponto-Caspian gammarids.

#### **Background**

Cleaning sampling equipment and fishing gear using high pressure water sprays may help to reduce the risk of transporting gammarid shrimps between water bodies.

### **3.1.11 Exchanging ballast water**

- No evidence was captured for exchanging ballast water to control Ponto-Caspian gammarids.

#### **Background**

Many freshwater invaders can be transported across oceans within the ballast water of ships. By exchanging ballast water collected at one freshwater port with salty ocean water during the journey, the chance of transporting live organisms to other freshwater ports is reduced. A study on the Laurentian Great Lakes has identified that possible non-compliance with a ballast exchange law prior to entering the lakes has facilitated repeated introductions of invasive shrimps (Hänfling *et al.* 2011).

Hänfling B., Edwards F. & Gherardi F. (2011) Invasive alien Crustacea: dispersal, establishment, impact and control. *BioControl*, 56, 573-595.

## 3.2 *Procambarus* crayfish

### Background

Swamp crayfish *Procambarus* spp. that have been introduced into European countries can survive prolonged periods out of water and can walk long distances over land to move between waterbodies. They are omnivorous, destroy bottom-rooting plants and feed heavily on insects and molluscs. They reduce resources available for native species. Swamp crayfish can be vectors for a number of parasites and can transmit crayfish plague to native European crayfish. The swamp crayfish itself is highly resistant to this disease.

### Key messages

#### ***Trapping and removal***

One controlled, replicated study from Italy found that food (tinned meat) was a more effective bait in trapping red swamp crayfish, than using pheromone treatments or no bait (control). Baiting with food increased trapping success compared to trapping without bait.

#### ***Encouraging predators***

Two replicated, controlled studies in Italy found that eels fed on the red swamp crayfish and reduced population size. One replicated, controlled study found that pike predated red swamp crayfish.

#### ***Trapping combined with encouragement of predators***

One before-and-after study from Switzerland and a replicated, paired site study from Italy found that a combination of trapping and predation was more effective at reducing red swamp crayfish populations than predation alone.

#### ***Sterilization of males***

One replicated laboratory study from Italy found that exposing male red swamp crayfish to X-rays reduced the number of offspring they produced.

#### ***Food source removal***

No evidence was captured on the effect of removing food sources as a control tool for *Procambarus* crayfish.

#### ***Draining the waterway***

No evidence was captured on the effect of draining the waterway as a control tool for *Procambarus* crayfish

#### ***Remove the crayfish by electrofishing***

No evidence was captured on the effect of electrofishing as a management control tool for *Procambarus* crayfish.

### **Add chemicals to the water**

One replicated study in Italy found that natural pyrethrum at concentrations of 0.05 mg/l and above was effective at killing red swamp crayfish both in the laboratory and in a river, but not in drained burrows.

### **Create barriers**

One before-and-after study from Italy found that the use of concrete dams across a stream was effective at containing spread of the population upstream.

### **Relocate vulnerable crayfish**

No evidence was captured for the effect of relocating native species as a management tool against the effects of *Procambarus* crayfish.

## **3.2.1 Trapping and removal**

- A controlled, replicated study in Italy<sup>1</sup> found that baiting traps with food (tinned meat) trapped the most red swamp crayfish compared to the use of male and female pheromones or the control (no bait). Over half of all crayfish caught were found in traps baited with food.

### **Background**

Traps are commercially available for crayfish capture. They are typically baited with food items and set overnight, after which crayfish are disposed of in a humane way. A number of studies have investigated the use of trapping to control other invasive crayfish species. A replicated study from the UK (Holdich & Black 2007) found that trapping spiny-cheeked crayfish *Orconectes limosus* was ineffective at managing populations. Another before and after study in Spain (Dana *et al.* 2010) demonstrated that a combination of trapping, manual removal and electrofishing controlled recruitment of signal crayfish *Pacifastacus leniusculus*. However, trapping contributed towards only approximately 3% of the total crayfish removed in this Spanish study.

Holdich D. & Black J. (2007). The spiny-cheek crayfish, *Orconectes limosus* (Rafinesque, 1817) [Crustacea:Decapoda:Cambaridae], digs into the UK. *Aquatic Invasions*, 2, 1-15.

Dana E.D., López-Santiago J., García-de-Lomas J., García-Ocaña D.M., Gámez V. & Ortega F. (2010). Long-term management of the invasive *Pacifastacus leniusculus* (Dana, 1852) in a small mountain stream. *Aquatic Invasions*, 5, 317-322.

A controlled, replicated study conducted in 2006 in canals in Italy<sup>1</sup> found that food-baited traps were successful in capturing red swamp crayfish *Procambarus clarkii*. Of 282 crayfish caught using different bait types, over half were captured in food-baited traps compared with traps containing male or female crayfish and a control treatment (no bait). A total of 72 traps were set three metres apart and randomly assigned one of four bait treatments: no bait, tinned meat (food), male crayfish, or female crayfish. Bait crayfish were kept inside a wire netting box inside the traps to prevent them from mating with trapped individuals. The traps were checked after two days. The sex of each trapped crayfish was determined.

(1) Aquiloni L. & Gherardi F. (2010) Crayfish females eavesdrop on fighting males and use smell and sight to recognize the identity of the winner. *Animal Behaviour*, 79, 265-269.

### 3.2.2 Encouraging predators

- Two replicated, controlled studies<sup>1</sup> in Italy found that eels fed on the red swamp crayfish and reduced population size.
- One replicated, controlled study<sup>2</sup> from France in 2001 found that pike predated red swamp crayfish.

#### Background

High levels of predation, especially from fish, may help to reduce crayfish population density. A study of red swamp crayfish introduced to Portugal (Correia 2001) found that mammal predators included red fox, otter, common genet and Egyptian mongoose but not weasel, polecat, stone marten, badger or wild cat. The study reported important bird predators as night heron and white stork, and to a lesser extent little egret, purple heron and grey heron.

Correia A.M. (2001). Seasonal and interspecific evaluation of predation by mammals and birds on the introduced red swamp crayfish *Procambarus clarkii* (Crustacea, Cambaridae) in a freshwater marsh (Portugal). *Journal of Zoology*, 255, 533-541.

One replicated, controlled laboratory study conducted in 2006 on specimens from wetlands and irrigation ditches in Italy<sup>1</sup> found that the eel *Anguilla anguilla*, preyed effectively on red swamp crayfish *Procambarus clarkii* compared with the control tank (no predator). Different sizes of crayfish were preyed upon at a similar rate (one crayfish every four days/eel). The number of dead crayfish in tanks increased when the crayfish were moulting. Crayfish were weighed and measured and placed within four aerated plastic tanks (100 cm diameter, 30 cm depth). Three tanks held 10 hard-shelled, male crayfish of one size class and an eel. There were three size classes in total. One tank had 10 crayfish but no eel (control). Over 14 days, the number and weight of crayfish preyed upon were recorded. Dead crayfish were replaced with live individuals and experiments were replicated five times.

A second replicated, controlled study conducted in 2007 in a canal in Italy<sup>1</sup> found that the eel *Anguilla anguilla*, preyed effectively on red swamp crayfish *Procambarus clarkii* compared with the control tank (no predator). The eels predated more on smaller than on larger crayfish. Crayfish were measured and placed in cages (50 x 50 x 200 cm; 2 mm mesh width) that were 3 m apart from each other. Each cage held five hard-shelled, male crayfish of one size class. Three size classes were used in total. In half of the cages, an adult eel was placed. The other cages were controls (no eel). Over 20 days, once a week, number and size of dead crayfish was recorded.

One replicated, controlled study in France<sup>2</sup> in 2001 found that pike *Esox lucius* predated red swamp crayfish. Pike of 40-50 cm could eat crayfish above 8 cm in length, although smaller pike tended to eat smaller crayfish. Predation remained

equally high when fish prey (rudd) were available as an alternative source. Experiments were conducted in experimental water enclosures measuring 3 x 5m placed within small ponds. Crayfish refuges were created from tree branches and pike refuges from floating sheets of polystyrene. Six water enclosures contained pike of 16-46 mm length. Four of these had small rudd as alternative food for the pike. Red swamp crayfish of wide size range were added *ad libitum* to each pond and predation allowed for 15 days. Two additional control enclosures included crayfish added in the absence of any pike. No mortality was observed. Crayfish mortality was measured by draining ponds and collecting the remaining crayfishes.

(1) Aquiloni L., Brusconi S., Cecchinelli E., Tricarico E., Mazza G., Paglianti A. & Gherardi F. (2010). Biological control of invasive populations of crayfish: The European eel (*Anguilla anguilla*) as a predator of *Procambarus clarkii*. *Biological Invasions*, 12, 3817-3824.

(2) Neveu A. (2001) Can resident carnivorous fishes slow down introduced alien crayfish spread? Efficacy of 3 fishes versus 2 crayfish species in experimental design. *Bulletin Francais De La Peche Pisciculture*. 361, 683-704

### **3.2.3 Trapping combined with encouragement of predators**

- A before-and-after study in Switzerland<sup>1</sup> found that introducing predators, combined with trapping significantly reduced red swamp crayfish populations in a pond. A second replicated, controlled study from Italy<sup>2</sup> demonstrated that trapping and predation in combination was more effective at reducing red swamp crayfish populations than predation alone.

#### **Background**

Combinations of management strategies may have additive or even complementary (synergistic) effects. A combination of trapping and removal alongside encouragement of predators has the potential to manage crayfish populations to low population densities.

A before-and-after study conducted between 1997 and 2001 in ponds in Switzerland<sup>1</sup> found that the introduction of natural predators in combination with trapping lowered numbers of the red swamp crayfish *Procambarus clarkii*. Over a period of four years, the number of crayfish fell from 10,000 to 1,000. Approximately 15,000 crayfish were caught in total. Each night, 0.7-3.4 crayfish were caught/trap. From 1997 to 1999, the natural predators, eel *Anguilla anguilla*, and pike *Esox Lucius*, were put in the pond along with 7,000 traps.

A replicated, paired sites study conducted in 2008 in two artificial canals in Italy<sup>2</sup> found that trapping was more effective than using a predator, the eel *Anguilla anguilla*, in controlling red swamp crayfish *Procambarus clarkii* populations. Transects containing low densities of eel did not effectively reduce red swamp crayfish *Procambarus clarkii* densities in comparison to transects containing traps.

When trapping was suspended for a month, crayfish populations increased, indicating that trapping effectively reduces crayfish population sizes. In each canal, two transects (150 x 3 m) were delimited using 2 mm-mesh wire netting. Crayfish density was estimated by trapping with 6 traps per transect for one week. After one week, 15 adult eels were put in one transect/canal. Trapping continued for two weeks. The sex and length of each trapped crayfish was determined. The number of live crayfish was monitored.

(1) Hefti D. & Stucki P. (2006) Crayfish management for Swiss waters. *Bulletin Francais De La Peche Pisciculture*, 380-81, 937-950.

(2) Aquiloni L., Brusconi S., Cecchinelli E., Tricarico E., Mazza G., Paglianti A. & Gherardi F. (2010). Biological control of invasive populations of crayfish: The European eel (*Anguilla anguilla*) as a predator of *Procambarus clarkii*. *Biological Invasions*, 12, 3817-3824.

### 3.2.4 Sterilisation of males

- A replicated laboratory study in Italy<sup>1</sup> found that exposing male red swamp crayfish to X-rays reduced the number of offspring they produced by 43%.

#### Background

Sterilisation of males which are allowed to remain in wild populations can help to reduce the rate of population increase. For example, a review study from the UK (Stebbing *et al.* 2012) reported that removal of pleopods used by males to deposit spermatophores onto the females led to a population decrease over a three year period.

Stebbing P.D., Longshaw M., Taylor N., Norman R., Lintott R., Pearce F. & Scott A. (2012) Review of methods for the control of invasive crayfish in Great Britain. *CEFAS. Contract C5471 final report*. 105 pp.

A replicated laboratory study conducted between 2005 and 2006 in Italy<sup>1</sup> found that male red swamp crayfish *Procambarus clarkii*, exposed to X-rays had a reduced reproductive ability. The number of offspring they successfully produced was reduced by 43% compared to a control group (no x-ray exposure). X-ray exposure did not affect the males' survival and mating abilities. Irradiated males had smaller testes and altered sperm production that lasted for at least a year. A total of 122 males were tested, half in a control group with no irradiation. Male crayfish were placed individually inside a plastic tube and exposed to a 6 MeV electron beam for five minutes. Testes and sperm production were measured, as were the number of viable offspring produced post-mating.

(1) Aquiloni L., Becciolini A., Berti R., Porciani S., Trunfio C. & Gherardi F. (2009), Managing invasive crayfish: use of X-ray sterilisation of males. *Freshwater Biology*, 54, 1510–1519.

(2)

### 3.2.5 Removal of food source

- No evidence was captured on the effect of removing food sources as a control tool for *Procambarus* crayfish.

#### Background

Crayfish are omnivores. Removing food items from their environment has the potential to reduce growth rates and survival and could lead to mortality. A replicated field study from California, USA (Pintor & Sih 2011), found that availability of prey, coupled with high levels of stream discharge limited the distribution of signal crayfish *Pacifastacus leniusculus* at transect and stream scales, respectively. A national study from the Czech Republic (Svobodová *et al.* 2012) found that invasive spiny-cheek crayfish *Oronectes limosus* were more commonly found in nutrient-enriched water while native species preferred water of a higher quality. In addition, a replicated study in Japan (Kobayashi *et al.* 2011) identified that abundant leaf litter content in ponds led to higher densities of red swamp crayfish, due to them using it as a food source. It suggested restricting leaf litter within ponds would reduce crayfish population sizes.

Pintor L.M. & Sih A. (2011). Scale dependent effects of native prey diversity, prey biomass and natural disturbance on the invasion success of an exotic predator. *Biological Invasions*, 13, 1357–1366.

Svobodová J., Douda K., Štambergováb M., Rí Pícek J., Vlach P. & Fischer D. (2012) The relationship between water quality and indigenous and alien crayfish distribution in the Czech Republic: patterns and conservation implications. *Aquatic conservation: marine and freshwater ecosystems*, 22, 776–786.

Kobayashi R., Maezono Y. & Miyashita T. (2011) The importance of allochthonous litter input on the biomass of an alien crayfish in farm ponds. *Population Ecology*, 53, 525-534.

### 3.2.6 Draining the waterway

- No evidence was captured on the effect of draining the waterway as a control tool for *Procambarus* crayfish.

#### Background

Draining a water body offers a potential control of invasive crayfish by making the location uninhabitable. While some individuals may die as a result of such management, many species are able to tolerate exposure to the air and can walk to adjacent water bodies. A trial in the Czech Republic found that three successive drawdowns of a 0.16 ha pond, followed by hand removal, was not able to remove all signal crayfish *Pacifastacus leniusculus* (Zozak & Polícar 2002). In addition, a review reported that draining habitat can have a significant, short-term impact, reducing populations of red swamp crayfish *Procambarus clarkia* (Stebbing *et al.* 2012).

However, it also reported that this approach causes significant impacts on natural habitats, is expensive, and does not lead to eradication.

Zozak P. & Policar, T. (2002) Practical elimination of signal crayfish, *Pacifastacus leniusculus* (Dana), from a pond. Pages 200-208 In: D.M. Holdich & P.J. Sibley (Eds.) *Management and Conservation of Crayfish* Proceedings of a conference held on 7th November 2002 at the Nottingham Forest Football Club, Nottingham, UK.

Stebbing P.D., Longshaw M., Taylor N., Norman R., Lintott R., Pearce F. & Scott A. (2012) Review of methods for the control of invasive crayfish in Great Britain. *CEFAS. Contract C5471 final report*. 105 pp.

### **3.2.7 Remove the crayfish by electrofishing**

- No evidence was captured on the effect of electrofishing as a control tool for *Procambarus* crayfish.

#### **Background**

Electric currents are often used for the management of freshwater fishes. Typically the fish are stunned and removed by nets. Crayfish are also susceptible to electric shocks. A study in the UK of a different species of invasive crayfish, the signal crayfish found that high intensity electric (96 kW, direct current 1600 V, 57.8 A, at 7 Hz) shocks delivered repeatedly via electrode tapes to two sections of stream resulted in crayfish mortality of 86-97% (Peay *et al.* 2014). All sizes of crayfish were affected, but small individuals (<30 mm carapace length) were more susceptible. Some crayfish survived in the stony banks. In addition, a review proposed that passing an electric current through water, using modified electrofishing gear, could kill the red swamp crayfish *Procambarus clarkia* (Stebbing *et al.* 2012). Employing standard electrofishing methods (with a boat-mounted electrode) stuns crayfish in open water which can then be collected by hand. Those in burrows will survive. A modified version of this equipment was developed for crayfish with a much higher current (96kW instead of 0.5kW of the normal set up, Stebbing *et al.* 2012). Electrofishing comes with inherent risks to the user and can only be safely conducted during summer months, in shallow, clear water.

Peay S., Dunn A.M., Kunin W.E., McKimm R. & Harrod C. (2014) A method test of the use of electric shock treatment to control invasive signal crayfish in streams. *Aquatic Conservation: Marine and Freshwater Ecosystems*, DOI: 10.1002/aqc.2541.

Stebbing P.D., Longshaw M., Taylor N., Norman R., Lintott R., Pearce F. & Scott A. (2012) Review of methods for the control of invasive crayfish in Great Britain. *CEFAS. Contract C5471 final report*. 105 pp.

### 3.2.8 Add chemicals to the water

- One replicated, controlled study in Italy<sup>1</sup> found that red swamp crayfish could be killed using the natural pyrethrum Pyblast at a concentration of 0.05 mg/l, but that application to drained crayfish burrows was not effective.

#### Background

Adding chemical toxicants to the water offers the potential for localised control of invasive crayfishes. Chemical 'dips' for fishing and sampling gear can also be an effective biosecurity method for preventing the transport of crayfish plague. Most chemicals will have an impact on non-target organisms, and so their use must be carefully considered before being dosed. In many instances, the use of chemicals may be subject to local or national approval by regulatory authorities. Two before and after studies from Norway (Sandodden & Johnsen 2010) and Scotland (Peay *et al.* 2006) demonstrated that the biocides cypermethin and pyblast, respectively, were both effective at eradicating a different species of invasive crayfish, signal crayfish. A further study using chlorinated lime was ineffective at controlling signal crayfish in the Czech Republic (Zozak & Policar 2002).

Sandodden, R., & Johnsen, S. I. (2010) Eradication of introduced signal crayfish *Pasifastacus leniusculus* using the pharmaceutical BETAMAX VET®. *Aquatic Invasions*, 5, 75-81.

Peay S., Hiley P.D., Collen P & Martin I. (2006) Biocide treatment of ponds in Scotland to eradicate signal crayfish. *Bulletin Français de la Pêche et de la Pisciculture*, 380-381, 1363-1379.

Zozak P. & Policar T. (2002) Practical elimination of signal crayfish, *Pacifastacus leniusculus* (Dana), from a pond. Pages 200-208 in: D.M. Holdich & P.J. Sibley (Eds.) *Management and Conservation of Crayfish* Proceedings of a conference held on 7th November 2002 at the Nottingham Forest Football Club, Nottingham, UK.

One replicated, controlled study in 2009 in Italy<sup>1</sup> found that natural pyrethrum (Pyblast) concentrations of 0.05mg/l and higher resulted in 100% mortality of red swamp crayfish *Procambarus clarkii* under laboratory conditions, 95% mortality when 0.05mg/l Pyblast was applied to a drainage channel, but no mortality following application of 0.05 mg/l Pyblast into active crayfish burrows. For the drainage channel study, two 50m-long upstream control transects were compared with two downstream transects treated with 0.05 mg/l Pyblast, and mortality of crayfish in each transect was monitored recorded every 24 h for 96 h. For the burrow study, a 0.3 l solution of 0.05 mg/l Pyblast was injected up to 2 m inside active crayfish burrows after the channel had been drained. Crayfish population changes were assessed by comparing capture rates in four replicated, baited traps before and after the treatment.

(1) Cecchinelli E., Aquiloni L, Maltagliati G, Orioli G, Tricarico E & Gherardi F (2012) Use of natural pyrethrum to control the red swamp crayfish *Procambarus clarkii* in a rural district of Italy. *Pest Management Science*, 68, 839-844.

### 3.2.9 Create barriers

- A before-and-after study conducted between 2007 and 2010 in Spain<sup>1</sup> found that the use of concrete dams across a stream, specifically designed with features to prevent red swamp crayfish from crawling over them, were effective at containing spread of the population upstream.

#### Background

Physical barriers have the potential to prevent the spread of invasive crayfish, especially upstream within rivers. A review reported that building physical barriers was an effective mechanism to halt, or at least delay, the natural movement of red swamp crayfish *Procambarus clarkii* (Stebbing *et al.* 2012). However, a before-and-after study (Hänfling *et al.* 2011) found that an erected barrier in the River Buaa at the border between Sweden and Norway did not prevent the migration of signal crayfish *Pacifastacus leniusculus* into the Norwegian part of the river.

Stebbing P.D., Longshaw M., Taylor N., Norman R., Lintott R., Pearce F. & Scott A. (2012) Review of methods for the control of invasive crayfish in Great Britain. *CEFAS. Contract C5471 final report*. 105 pp.

Hänfling B., Edwards F. & Gherardi F. (2011) Invasive alien Crustacea: dispersal, establishment, impact and control. *BioControl*, 56, 573–595.

A before-and-after study from 2007 to 2010 in a mountain stream in Italy<sup>1</sup> found that building a series of small dams stopped migration of the red swamp crayfish *Procambarus clarkii*. The invasive crayfish did not penetrate into previously uninhabited areas upstream beyond the lower dams. In addition, numbers dropped below detectable levels in previously occupied areas in the mid reaches between dams. Dams were 1.5–2 m high and 6 m wide and constructed from reinforced concrete. Several design features discouraged crayfish from climbing over including vertical walls, smooth mortar, vertical wing-walls 3.5 m along the banks, and a projecting rim (crayfish are unable to walk upside down on a smooth surface). A flat stony platform was built downstream of each dam to create a shallow area with no refuges, discouraging crayfish from lingering near the dam. Crayfish populations were monitored for 30 days between July and October each year.

(1) Dana E.D., García-de-Lomas J., González R. & Ortega F. (2011) Effectiveness of dam construction to contain the invasive crayfish *Procambarus clarkii* in a Mediterranean mountain stream. *Ecological Engineering* 37, 1607–1613.

### 3.2.10 Relocate vulnerable native crayfish

- No evidence was captured for the effect of relocating native species as a management tool against the effects of *Procambarus* crayfish.

## Background

One of the greatest concerns about invasive crayfishes is their potential to displace native crayfish species, through transmission of crayfish plague or through competitive displacement. A randomised, replicated, controlled, before and after study from the UK (Haddaway *et al.* 2012) revealed that relocation of a native crayfish *Austropotamobius pallipes* to avoid the invasive signal crayfish, had no negative effects on growth, survival or morphology of the native species.

Haddaway N. R., Mortimer R.J.G., Christmas M., Grahame J.W., & Dunn A.M. (2012) Morphological diversity and phenotypic plasticity in the threatened British white-clawed crayfish (*Austropotamobius pallipes*). *Aquatic conservation: marine and freshwater ecosystems*, 22, 220–231.

## 4 Invasive fish

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### 4.1 Brown and black bullheads

#### Background

The brown bullhead *Ameiurus nebulosus* and black bullhead *Ameiurus melas* are native to North America. These small catfish grow to only 20cm, but can reach huge densities. They are tolerant of poor water quality and able to survive temperatures up to 35°C with low oxygen (Scott & Crossman 1973). They have an omnivorous diet which includes invertebrates, small vertebrates and fish eggs (Scott & Crossman 1973). Their strong dorsal and pectoral fin spines protect them from predators.

Introductions of these species may lead to competition for food or space and, combined with their predation on small native fishes and invertebrates, may affect ecosystem functioning and food webs. The feeding behaviour of brown and black bullheads may disturb bottom sediments and increase turbidity of the water. Black bullheads have previously established in the UK but were successfully eradicated through the use of the fish toxin rotenone.

Scott W.B. & Crossman E.J. (1973) Freshwater fishes of Canada. *Fisheries Research Board of Canada Bulletin*, 184, 966 pp.

#### Key messages

##### **Biological control using native predators**

No evidence was captured on the impact of native predators on invasive bullhead populations.

##### **Biological control of beneficial species**

No evidence was captured for reducing or controlling bullhead population size by reducing the population of co-occurring beneficial species.

##### **Application of a biocide**

Two studies in the UK and USA found that rotenone successfully eradicated black bullhead.

##### **Habitat manipulation**

No evidence was captured on the impact of habitat manipulation on invasive bullhead populations.

##### **Draining invaded waterbodies**

No evidence was captured for the use of draining invaded waterbodies to reduce the population size of invasive bullheads.

##### **Netting**

A replicated study in a nature reserve in Belgium found that double fyke nets could be used to significantly reduce the population of large brown bullheads.

### ***Electrofishing***

No evidence was captured for use of electrofishing to reduce the population size of invasive bullheads.

### ***Using a combination of netting and electrofishing***

No evidence was captured on the impact of electrofishing and gill netting combined on bullhead populations.

### ***Trapping using sound or pheromonal lures***

No evidence was captured for the effectiveness of trapping bullheads using sound or pheromonal lures.

### ***Increasing carbon dioxide concentrations***

No evidence was captured on the use of carbon dioxide for management of invasive bullheads.

### ***UV radiation***

No evidence was captured on the impact of UV radiation on bullhead populations.

### ***Changing salinity***

No evidence was captured on the impact of changing salinity on bullhead populations.

### ***Changing pH***

No evidence was captured on the impact of altering pH on bullhead populations.

### ***Public education***

No evidence was captured on the impact of education programmes on invasive bullhead populations.

## **4.1.1 Biological control using native predators**

- No evidence was found on the impact of native predators on invasive bullhead populations.

### **Background**

Encouraging native predators can potentially increase predation on the population of invasive bullheads, thereby biologically controlling bullhead populations.

Large game fish such as bass *Micropterus salmoides*, pike *Esox lucius*, pickerel *Esox niger*, and perch *Perca flavescens*, in addition to snapping turtles *Chelydra serpentina*, water snakes, and wading birds are known to prey upon brown bullheads (EPA 2015). In addition, it is reported that parasitic trematodes, cestodes, copepods and nematodes have been found in brown bullheads, though it is not clear to what extent they affect bullhead survival (EPA 2015).

Protective spines on bullheads and the species' preference for eating mostly at night, make bullheads an uncommon prey for other fish (Sigurdson 2012). However, pike, turtles, flathead catfish *Pylodictis olivaris*, great blue herons *Ardea herodias* and otters (subfamily Lutrinae) eat small bullheads up to four inches long.

It is considered that biological control of adult brown bullheads is unlikely given the paucity of natural predators within the native range, although juveniles may be predated upon by certain large-bodied fishes in the native range, such as *Esox* species (CABI 2015).

CABI (2015) Datasheet on *Ameiurus nebulosus* (brown bullhead). CABI Invasive Species Compendium. CABI International, Wallingford, UK. 21 pp.

EPA (2015) Brown Bullhead. Econorisk Profile. United States Environmental Protection Agency, Washington D.C., USA. 6pp.

Sigurdson R. (2012) Species profile – Bullheads. Minnesota Department of Natural Resources, USA. 2pp.

#### **4.1.2 Biological control of beneficial species**

- No evidence was found for reducing or controlling bullhead population size by reducing the population of co-occurring beneficial species.

##### **Background**

Numerous species benefit from the presence of beneficial species. Reducing the population of co-occurring beneficial species in localised populations, and limiting their spread or intentional introduction, may therefore offer a tool for managing and reducing bullhead populations.

#### **4.1.3 Application of a biocide**

- A study in the UK<sup>1</sup> reported that use of a piscicide containing rotenone achieved eradication of black bullhead.
- A study in the USA<sup>2</sup> found that rotenone successfully eradicated black bullhead, but one of two ponds required two separate doses.

##### **Background**

Biocides may offer a tool for localised eradication or population reduction of bullheads, provided potentially negative effects on native species are carefully managed.

Rotenone has been successfully used to control populations of bullheads and other fish. For example, in the UK, studies have reported that rotenone successfully reduced or eradicated populations of topmouth gudgeon *Pseudorasbora parva* and fathead minnows *Pimephales promelas*, from ponds and lakes (Britton *et al.* 2008; Britton *et al.* 2011). A risk assessment for piscicidal formulations of rotenone suggests that mortality of bullheads can be achieved with 5-100 parts per billion of

rotenone active ingredient, or 100-200 parts per billion of rotenone active ingredient in organic rich ponds, diluted in 38 litres of water, although no scientific evidence was provided (Turner *et al.* 2007).

It has been suggested that Antimycin is less useful for control of bullheads than rotenone as Antimycin is very toxic to scaled fishes, but is much less toxic to scaleless catfishes (Order Siluriformes), which includes the bullheads (Clearwater *et al.* 2008).

Britton R., Brazier M., Davies G.D. & Chare S.I. (2008) Case studies on eradicating the Asiatic cyprinid *Pseudorasbora parva* from fishing lakes in England to prevent their riverine dispersal. *Aquatic Conservation: Marine and Freshwater Ecosystems*, 18, 867-876.  
Britton R., Copp G.H., Brazier M. & Davies G.D. (2011) A modular assessment tool for managing introduced fishes according to risks of species and their populations, and impacts of management actions. *Biological Invasions*, 13, 2847-2860.  
Clearwater S.J., Hickey C.W. & Martin M.L. (2008) Overview of potential piscicides and molluscicides for controlling aquatic pest species in New Zealand. *Science for Conservation*, 283, 1-74.  
Turner L., Jacobson S., & Shoemaker L. (2007) *Risk Assessment for Piscicidal Formulations of Rotenone*. Compliance Services International, Washington, USA. 104pp.

A study in 2014 at a fishery in Essex, UK<sup>1</sup> reported that use of a piscicide containing rotenone achieved eradication of black bullhead *Ameiurus melas*. The piscicide was applied using a boat and a bank based application system. Dead fish were removed using nets. During and after the operation, regular water samples were taken to monitor the level of rotenone.

A study from 2001-2003 in two ponds in Illinois, USA<sup>2</sup> found that rotenone successfully eradicated black bullhead *Ameiurus melas*, but one pond required two separate doses due to an incomplete initial kill. Rotenone was applied in December 2001 using a motorised and hand-pumped sprayer at concentrations of 7 parts per million or 3.5 parts per million, with dose dependent on apparent fish susceptibility. It was applied from several points along banks to ensure complete coverage. A second application was applied in January 2003 as black bullhead catfish were not eliminated in 2001. Ponds were sampled with wire minnow traps, D-frame nets and visual observations to ensure fish had been eliminated.

(1) Environment Agency Invasive Species Action Group. (2014) Non-Native Species Newsletter: Spring Edition. Environment Agency Invasive Species Action Group report. 4pp.

(2) Towey J.B. (2007) *Influence of fish presence and removal on woodland pond breeding amphibians*. MSc thesis. Eastern Illinois University.

#### **4.1.4 Habitat manipulation**

- No evidence was captured on the impact of habitat manipulation on invasive bullhead populations.

#### **Background**

Habitat manipulation, such as removing protective vegetation cover, can offer a technique for altering species abundance within discreet environments.

#### 4.1.5 Draining invaded waterbodies

- No evidence was found for use of draining invaded waterbodies to reduce the population size of invasive bullheads.

##### Background

Draining invaded waterbodies is an effective tool for reducing the population size of other fish species such as topmouth gudgeon *Pseudorasbora parva*, potentially used in combination with gill netting and electrofishing (Copp *et al.* 2007), or other habitat modifications such as pH alteration (Britton *et al.* 2008). It is therefore possible that it could also prove a useful tool for reducing populations of invasive bullheads.

Potential negative side effects of draining waterbodies on native species need to be carefully managed. However, it is possible that eradicating invasive bullheads from a waterbody would facilitate an increase in the diversity and richness of native species, as found when invasive largemouth bass *Micropterus salmoides* were eradicated through pond drying in a field study in Japan (Tsunoda *et al.* 2010).

Britton R., Brazier M., Davies G.D. & Chare S.I. (2008) Case studies on eradicating the Asiatic cyprinid *Pseudorasbora parva* from fishing lakes in England to prevent their riverine dispersal. *Aquatic Conservation: Marine and Freshwater Ecosystems*, 18, 867-876.

Copp G.H., Wesley K.J., Verreycken H. & Russell I.C. (2007) When an 'invasive' fish species fails to invade! Example of the topmouth gudgeon *Pseudorasbora parva*. *Aquatic Invasions*, 2, 107-112.

Tsunoda H., Mitsuo O., Ohira M., Doi M. & Senga Y (2010). Change of fish fauna in ponds after eradication of invasive piscivorous largemouth bass, *Micropterus salmoides*, in north-eastern Japan. *Aquatic Conservation: Marine and Freshwater Ecosystems*, 20, 710-716.

#### 4.1.6 Netting

- A replicated study from 1999-2000 in shallow interconnected ponds in a nature reserve in Belgium<sup>1</sup> found that double fyke nets could be used to significantly reduce the population of brown bullhead measuring over 8cm

##### Background

Netting provides a potential tool for localised population reduction of invasive bullheads

A replicated study from 1999-2000 conducted in small, shallow, interconnected ponds in a nature reserve in Belgium<sup>1</sup>, found that double fyke nets were an effective tool to significantly reduce the population of brown bullhead *Ameiurus nebulosus*. In ponds smaller than 1.5 hectares, a set of 12-16 double fyke nets caught an average of 66% (maximum 80%) of all brown bullhead measuring over 8 cm within one to two days. Capture rates were lower in a larger pond of three hectares. Mean depth

of the ponds was 1.5 meters. The fyke nets consisted of two conically shaped nets connected by a vertically hanging net (11 x 1 m). Each fyke net had a total length of 8 m and mesh size of 8 mm. Trapped fish within the nets were counted and measured over one to two days.

(1)Louette G. & Declerk S. (2006) Assessment and control of non-indigenous brown bullhead *Ameiurus nebulosus* populations using fyke nets in shallow ponds. *Journal of Fish Biology*, 68, 522-531.

#### **4.1.7 Electrofishing**

- No evidence was found for use of electrofishing to reduce the population size of invasive bullheads.

##### **Background**

Electrofishing provides a potential, although non-selective, tool for localised population reduction of invasive bullheads.

#### **4.1.8 Using a combination of netting and electrofishing**

- No evidence was captured on the impact of electrofishing and gill netting combined on bullhead populations.

##### **Background**

Applying a combination of netting and electrofishing may prove particularly effective in controlling populations of invasive bullheads. Two before-and-after studies from Australia (Pinto *et al.* 2005) and the UK (Copp *et al.* 2007), found that electrofishing and gill netting combined were effective at reducing carp *Cyprinus carpio* and topmouth gudgeon *Pseudorasbora parva* populations. A replicated, paired sites study in the USA demonstrated that gill netting and electrofishing reduced or eradicated non-native trout species and facilitated a partial reversal in decline of the yellow-legged frog *Rana muscosa* (Knapp *et al.* 2007).

Copp G.H., Wesley K.J., Verreycken H. & Russell I.C. (2007) When an 'invasive' fish species fails to invade! Example of the topmouth gudgeon *Pseudorasbora parva*. *Aquatic Invasions*, 2, 107-112.

Knapp R.A., Boiano D.M. & Vredenburg V.T. (2007) Removal of nonnative fish results in population expansion of a declining amphibian (mountain yellow-legged frog, *Rana muscosa*). *Biological Conservation*, 135, 11-20.

Pinto L., Chandrasena N., Pera J., Hawkins P., Eccles D. & Sim R. (2005) Managing invasive carp (*Cyprinus carpio* L.) for habitat enhancement at Botany Wetlands, Australia. *Aquatic Conservation: Marine and Freshwater Ecosystems*, 15, 447-462.

#### 4.1.9 Trapping using sound or pheromonal lures

- No evidence has been found for the effectiveness of trapping bullheads using sound or pheromonal lures.

##### Background

Brown bullheads are notable for their sound production. In the lab, they produce sound during aggressive, conspecific encounters (Anderson et al. 2008). Certain fish species, such as the round goby *Neogobius melanostomus*, are also attracted to pheromones from the same species (Gammon et al. 2005). It is possible that there is potential for trapping bullhead fish using sound or pheromonal lures provided that possible influences on non-target species are addressed.

Anderson, K., Rountree, R. & Juanes, F. (2008) Soniferous fishes in the Hudson River. *Transactions of the American Fisheries Society*, 137, 616-626.

Gammon D.B., Li W., Scott A.P., Zielinski B.S. & Corkum L.D. (2005) Behavioural responses of female *Neogobius melanostomus* to odours of conspecifics. *Journal of Fish Biology*, 67, 615-626.

#### 4.1.10 Increasing carbon dioxide concentrations

- No evidence was captured on the use of carbon dioxide for management of invasive bullheads.

##### Background

Addition of carbon dioxide to water in the form of dry ice displaces dissolved oxygen in the water, thereby decreasing its concentration. The increased carbon dioxide concentration has a weakly anaesthetic action on fishes (Clearwater et al. 2008).

Increasing carbon dioxide concentrations by either bubbling the pressurised gas directly into water, or by the addition of sodium bicarbonate has been used to sedate fishes during transport or to allow handling of large numbers of fishes, with minimal residual toxicity (Clearwater et al. 2008). A previous study found that exposure to sodium bicarbonate at a concentration of 142–642 mg/L for 5 min can anaesthetise some fish species (Brooke et al. 1978).

Addition of dry ice may therefore offer a tool enabling selective removal of invasive bullheads from waterbodies. This is provided that sufficiently high carbon dioxide levels can be maintained, which may be difficult, particularly in natural water bodies. Also, bullhead fish can tolerate relatively low ambient oxygen levels, although it is possible that rapid reduction in oxygen levels may have a sedative effect.

Brooke H.E., Hollender, B. & Lutterbie G. (1978) Sodium bicarbonate, an inexpensive fish anesthetic for field use. *The Progressive Fish-Culturist*, 40, 11–13.

Clearwater S.J., Hickey C.W. & Martin M.L. (2008) Overview of potential piscicides and molluscicides for controlling aquatic pest species in New Zealand. *Science for Conservation*, 283, 1-74.

#### **4.1.11 UV radiation**

- No evidence was captured on the impact of UV radiation on bullhead populations.

##### **Background**

Applying UV radiation to an invaded waterbody may offer a tool for localised eradication or population reduction of bullheads, provided potentially negative effects on native species are carefully managed. A combined laboratory and field study in Lake Tahoe, USA indicated that UV levels in parts of the lake are intense enough to kill all larvae of largemouth bass *Micropterus salmoides* (Tucker *et al.* 2012).

Tucker A.J., Williamson C.E. & Oris J.T. (2012) Development and application of a UV attainment threshold for the prevention of warm water aquatic invasive species. *Biological Invasions*, 14, 2331-2342.

#### **4.1.12 Changing salinity**

- No evidence was captured on the impact of changing salinity on bullhead populations.

##### **Background**

Changing the salinity of an invaded waterbody may offer a tool for localised eradication or population reduction of bullheads, provided potentially negative effects on native species are managed carefully.

#### **4.1.13 Changing pH**

- No evidence was captured on the impact of altering pH on bullhead populations.

##### **Background**

Some species of fish are sensitive to pH changes and this can offer a tool for localised eradication or population reduction, provided potentially negative effects on native species are carefully managed. Draining ponds and altering pH has been shown to be effective in local eradication of other fish species. For example, a before-and-

after study in the UK found that draining ponds and altering pH using lime slurry, eradicated topmouth gudgeon *Pseudorasbora parva* (Britton *et al.* 2008).

Britton R., Brazier M., Davies G.D. & Chare S.I. (2008) Case studies on eradicating the Asiatic cyprinid *Pseudorasbora parva* from fishing lakes in England to prevent their riverine dispersal. *Aquatic Conservation: Marine and Freshwater Ecosystems*, 18, 867-876.

#### **4.1.14 Public education**

- No evidence was captured on the impact of education programmes on invasive bullhead populations.

#### **Background**

Public education programmes on the risks of invasive bullheads and the mechanisms of spread could reduce the translocation of bullheads into new water bodies, thereby reducing population spread.

## 4.2 Ponto-Caspian gobies

### Background

The round goby *Neogobius melanostomus* and tubenose goby *Proterorhinus marmoratus*, have been some of the most successful fish invaders in recent decades, with invasive populations established widely across Western Europe and North America.

Gobies are bottom-feeding omnivores, consuming a wide variety of benthic invertebrates, such as chironomids, crustaceans, copepods, dipterans, ephemeropterans, ostracods, and trichopterans, and occasionally larval fishes. Gobies may have dietary overlap with some native fish species and so may compete with them for food.

The invasion of the round goby in the Baltic Sea has resulted in diet shift among predators and changes in the structures of food webs (Janssen & Jude 2001). In the Great Lakes of North America some native fish species have declined due to failure to recruit following the establishment of round gobies (Corkum *et al.* 2004).

Ponto-Caspian gobies feed predominantly on molluscs such as zebra mussels *Dreissena polymorpha* and quagga mussels *Dreissena bugensis*, which has the potential to control other Ponto-Caspian invaders. However, zebra and quagga mussels can accumulate toxins such as heavy metals and organochlorines from the environment, which may result higher concentrations of these pollutants higher up the food chain. This may have harmful effects on top predators and the fish diet of humans is a health concern (Charlebois *et al.* 1997).

Charlebois P.M., Marsden J.E., Goettel R.G., Wolf R.K., Jude D.J. & Rudnika S. (1997) The round goby *Neogobius melanostomus* (Pallas, 1811), a review of European and North American Literature. *Illinois Natural History Survey Special Publication*, 1-76.

Corkum L.D., Sapota M.R. & Skora K.E. (2004) The round goby, *Neogobius melanostomus*, a fish invader on both sides of the Atlantic Ocean. *Biological Invasions*, 6, 173-181.

Janssen J. & Jude D.J. (2001) Recruitment failure of mottled sculpin *Cottus bairdi* in Calumet Harbor, southern Lake Michigan, induced by the newly introduced round goby *Neogobius melanostomus*. *Journal of Great Lakes Research*, 27, 319-328.

### Key messages

#### ***Biological control using native predators***

No evidence was captured on the deliberate introduction of a native predator to biologically control gobies.

#### ***Biological control of beneficial species***

No evidence was captured for reducing or controlling goby population size by reducing the population of co-occurring beneficial species.

***Application of a biocide***

No evidence was captured on the use of biocide to control populations of the round goby or the tubenose goby.

***Habitat manipulation***

No evidence was captured on the use of habitat manipulation to control invasive goby populations.

***Draining invaded waterbodies***

No evidence was captured for the use of draining waterbodies to reduce the population size of invasive gobies.

***Netting***

No evidence was captured on seine netting to reduce the size of goby populations.

***Electrofishing***

No evidence was captured for use of electrofishing to reduce the population size of invasive gobies.

***Using a combination of netting and electrofishing***

No evidence was captured on the use of a combination of electrofishing and gill netting to control goby populations.

***Trapping using visual, sound and pheromonal lures***

No evidence was captured for the effectiveness of trapping Ponto-Caspian gobies using visual, sound or pheromonal lures.

***Increasing carbon dioxide concentrations***

No evidence was captured on the use of carbon dioxide as a control measure for Ponto-Caspian gobies.

***UV radiation***

No evidence was captured on the use of UV radiation to control goby populations.

***Changing salinity***

A replicated controlled laboratory study in Canada found 100% mortality of round gobies within 48 hours of exposure to water of 30% salinity.

***Changing pH***

No evidence was captured on the use of pH alteration to control goby populations.

***Use of barriers to prevent migration***

A controlled, replicated field study in the USA found that an electrical barrier prevented movement of round gobies across it, and that increasing electrical pulse duration and voltage increased the effectiveness of the barrier.

***Public education***

No evidence was captured on the impact of education programmes on control of goby populations.

**4.2.1 Biological control using native predators**

- No evidence was captured on the deliberate introduction of a native predator to biologically control gobies.

## Background

Encouraging native predators could potentially increase predation on the population of invasive gobies, thereby biologically controlling goby populations. For example, a before-and-after field and modelling study, from 1999-2008 in Lake Erie, North America suggested that predator control of round goby *Neogobius melanostomus*, by native burbot *Lota lota* could be effective at controlling population size (Madenjian *et al.* 2011). Populations of native burbot were reported to have high potential for predatory control of invasive round gobies and computer modelling indicated that in 2007, burbot consumed approximately 61% of round goby standing stock (Madenjian *et al.* 2011). Although many species consume round goby, no effective and species-specific biocontrol has yet been reported. More work is therefore required.

Madenjian C.P., Stapanian M.A., Witzel L.D., Einhouse D.W., Pothoven S.A. & Whitford H.L. (2011) Evidence for predatory control of the invasive round goby. *Biological Invasions*, 13, 987-1002.

### 4.2.2 Biological control of beneficial species

- No evidence was found for reducing or controlling goby population size by reducing the population of co-occurring beneficial species.

## Background

Numerous species benefit from the presence of beneficial species. Reducing the population of co-occurring beneficial species in localised populations, and limiting their spread or intentional introduction, may therefore offer a tool for managing and reducing goby populations.

### 4.2.3 Application of a biocide

- No evidence was captured on the use of biocide to control populations of the round goby or the tubenose goby.

## Background

Biocides may offer a tool for localised eradication or population reduction of gobies, provided potentially negative effects on native species are carefully managed.

Whilst a number of different biocides may be considered for control of goby populations, no evidence has been found relating to detailed scientific study of any of these biocides to control an existing population. For example, delayed-release and/or pelleted formulations of Bayluscide® are said to have been used in the USA to

selectively reduce populations of bottom-associated fish species such as the round goby *Neogobius melanostomus* (Clearwater *et al.* 2008). It is reported that the round goby is unable to detect Bayluscide® and that exposure for a few minutes is lethal to the round goby, even if the fishes are removed to freshwater immediately afterwards (Schreier *et al.* 2001). Of the four chemical piscicides registered for use in the USA, rotenone and antimycin A and are considered “general” piscicides, but no studies have been found of their effects on round goby. Studies have however been conducted on the effects of rotenone on other fish species. For example, in the UK, before-and-after studies have reported that rotenone successfully reduced or eradicated populations of topmouth gudgeon *Pseudorasbora parva* and fathead minnows *Pimephales promelas* from ponds and lakes (Britton *et al.* 2008; Britton *et al.* 2011).

The effect of saponins on other fish species has also been studied. For example, a study in New Zealand found that an increase in water temperature or a decrease in dissolved oxygen concentration increases the sensitivity of invasive African flathead goby *Glossogobius giurus* to saponins provided in the form of teaseed cake (Minsalan & Chiu 1986). African flathead goby were eliminated by an application of 15 mg/L teaseed cake (Minsalan & Chiu 1986). However, no evidence was captured on the impact of saponins on the round goby or the tubenose goby *Proterorhinus marmoratus*.

Britton R., Brazier M., Davies G.D. & Chare S.I. (2008) Case studies on eradicating the Asiatic cyprinid *Pseudorasbora parva* from fishing lakes in England to prevent their riverine dispersal. *Aquatic Conservation: Marine and Freshwater Ecosystems*, 18, 867-876.

Britton R., Copp G.H., Brazier M. & Davies G.D. (2011) A modular assessment tool for managing introduced fishes according to risks of species and their populations, and impacts of management actions. *Biological Invasions*, 13, 2847-2860.

Clearwater S.J., Hickey C.W. & Martin M.L. (2008) Overview of potential piscicides and molluscicides for controlling aquatic pest species in New Zealand. *Science for Conservation*, 283, 1-74.

Minsalan C.L.O. & Chiu Y.N. (1986) Effects of teaseed cake on selective elimination of finfish in shrimp ponds. Pp. 79–82 In Maclean J.L., Dizon L.B. & Hosillos, L.V. (Eds) *The First Asian Fisheries Forum. Proceedings of the First Asian Fisheries Forum*, Manila, Philippines, 26–31 May 1986. pp. 79-82.

Schreier T.M., Dawson V.K., Larson W.J. & Schleis S.M. (2001) Piscicides as an emergency tool for controlling range expansion of the round goby (*Neogobius melanostomus*) in the Illinois waterway. Abstracts from the 44th Conference on Great Lakes Research, June 10–14, 2001, Green Bay, IL. International Association of Great Lakes Research, Ann Arbor, MI. p.120

#### **4.2.4 Habitat manipulation**

- No evidence was captured on the use of habitat manipulation to control invasive goby populations.

#### **Background**

Habitat manipulation, such as removing protective cover from vegetation, can offer a technique for altering species abundance within discreet environments.

#### 4.2.5 Draining invaded waterbodies

- No evidence was captured for use of draining waterbodies to reduce the population size of invasive gobies.

##### Background

Draining invaded waterbodies is a proven tool for reducing the population size of fish such as topmouth gudgeon *Pseudorasbora parva*, potentially used in combination with gill netting and electrofishing (Copp *et al.* 2007), or other habitat modifications such as pH alteration (Britton *et al.* 2008). It is therefore possible that it could also prove a useful tool for reducing populations of invasive gobies.

Potentially negative side effects of draining waterbodies on native species need to be carefully managed. However, it is possible that eradicating invasive gobies from a waterbody would facilitate an increase in the diversity and richness of native species, as found when invasive largemouth bass *Micropterus salmoides* were eradicated through pond drying in a field study in Japan (Tsunoda *et al.* 2010).

Britton R., Brazier M., Davies G.D. & Chare S.I. (2008) Case studies on eradicating the Asiatic cyprinid *Pseudorasbora parva* from fishing lakes in England to prevent their riverine dispersal. *Aquatic Conservation: Marine and Freshwater Ecosystems*, 18, 867-876.

Copp G.H., Wesley K.J., Verreycken H. & Russell I.C. (2007) When an 'invasive' fish species fails to invade! Example of the topmouth gudgeon *Pseudorasbora parva*. *Aquatic Invasions*, 2, 107-112.

Tsunoda H., Mitsuo O., Ohira M., Doi M. & Senga Y (2010). Change of fish fauna in ponds after eradication of invasive piscivorous largemouth bass, *Micropterus salmoides*, in north-eastern Japan. *Aquatic Conservation: Marine and Freshwater Ecosystems*, 20, 710-716.

#### 4.2.6 Netting

- No evidence was captured on the use of seine netting to control goby populations.

##### Background

Gill netting and seine netting provide a potential tool for localised population reduction of invasive gobies. Studies have shown this to be effective for other fish species. For example, a before-and-after study in the UK found repeated removal of topmouth gudgeon *Pseudorasbora parva* by seine netting reduced pond and lake populations (Britton *et al.* 2010).

Britton J. R., Davies G.D. & Brazier M. (2010) Towards the successful control of the invasive *Pseudorasbora parva* in the UK. *Biological Invasions*, 10, 125-131.

#### 4.2.7 Electrofishing

- No evidence was captured for use of electrofishing to reduce the population size of invasive gobies.

##### Background

Electrofishing provides a potential, although non-selective, tool for localised population reduction of invasive gobies.

#### 4.2.8 Using a combination of netting and electrofishing

- No evidence was captured on the use of a combination of electrofishing and gill netting to control goby populations.

##### Background

Applying a combination of netting and electrofishing may prove particularly effective in controlling populations of invasive gobies. Studies have shown this to be effective for other fish species. For example, two before-and-after studies in Australia and the UK found that electrofishing and gill netting combined were effective at reducing topmouth gudgeon *Pseudorasbora parva* populations (Pinto *et al.* 2005; Copp *et al.* 2007). Also, a replicated, paired sites study in the USA demonstrated that gill netting and electrofishing reduced or eradicated non-native trout (*Oncorhynchus* species, *Salmo* species, and *Salvelinus* species), and facilitated a partial reversal in the decline in yellow-legged frog *Rana muscosa* (Knapp *et al.* 2007).

Copp G.H., Wesley K.J., Verreycken H. & Russell I.C. (2007) When an 'invasive' fish species fails to invade! Example of the topmouth gudgeon *Pseudorasbora parva*. *Aquatic Invasions*, 2, 107-112.

Knapp R.A., Boiano D.M. & Vredenburg V.T. (2007) Removal of nonnative fish results in population expansion of a declining amphibian (mountain yellow-legged frog, *Rana muscosa*). *Biological Conservation*, 135, 11-20.

Pinto L., Chandrasena N., Pera J., Hawkins P., Eccles D. & Sim R. (2005) Managing invasive carp (*Cyprinus carpio* L.) for habitat enhancement at Botany Wetlands, Australia. *Aquatic Conservation: Marine and Freshwater Ecosystems*, 15, 447-462.

#### 4.2.9 Trapping using visual, sound and pheromonal lures

- No evidence was captured on trapping Ponto-Caspian gobies using sound or pheromonal lures.

## Background

Certain fish species, such as the round goby *Neogobius melanostomus*, are attracted to pheromones of the same species (Gammon *et al.* 2005). This indicates the potential for trapping using sound and pheromonal lures, provided that possible influences on non-target species are addressed. However, whilst a number of studies have identified sound and pheromonal lures, no evidence has been found for the effectiveness of trapping round gobies or tubenose gobies *Pseudorasbora parva* using such lures.

Studies have shown the nature of pheromonal attraction in gobies. For example, two controlled studies demonstrated that reproductive female round gobies were attracted to reproductive male goby pheromones, whilst immature females were attracted to reproducing females but not the male pheromones (Gammon *et al.* 2005; Corkum *et al.* 2006). Also, a controlled study on well-fed juvenile round gobies found that they were attracted to the odour of eggs from their own species (Yavno & Corkum 2011).

Other studies have shown the nature of sound attraction. For example, a replicated, controlled experiment in the laboratory and in the field in the USA found that round gobies showed a highly directed response to playbacks of the calls of the same species (Rollo *et al.* 2007). Female round gobies in particular, showed significant attraction to speakers emitting same species male calls.

Other studies have shown the nature of visual attraction. For example, a controlled study determined that mature female round gobies were attracted to plastic models of male fish, specifically dark (reproductive) rather than mottled (immature) models (Yavno & Corkum 2009).

It may be that a combined approach of visual, sound and pheromonal lures will lead to the highest capture rate. However, it will be important to identify lures that are specific to invasive gobies. Some research leads to species-specific lures. For example, a field study in the USA reported that amino acids and bile acid released by reproductive round gobies consistently resulted in electrical activity in the smell receptors of five other species tested, but only round gobies showed a response to pheromones produced by reproductive male gobies indicating that the pheromones were species-specific in this instance (Ochs *et al.* 2013).

Corkum L.D., Arbuckle W.J., Belanger A.J., Gammon D.B., Weiming L. Scott A.P. & Zielinski B (2006) Evidence of a male sex pheromone in the round goby (*Neogobius melanostomus*). *Biological Invasions*, 8, 105-112.

Gammon D.B., Li W., Scott A.P., Zielinski B.S. & Corkum L.D. (2005) Behavioural responses of female *Neogobius melanostomus* to odours of conspecifics. *Journal of Fish Biology*, 67, 615-626.

Ochs C.L., Laframboise A.J., Green W.W., Basilius A., Johnson T.B. & Zielinski B.S. (2013) Response to putative round goby (*Neogobius melanostomus*) pheromones by centrarchid and percid fish species in the Laurentian Great Lakes. *Journal of Great Lakes Research*, 39, 186-189.

Rollo A., Andraso G., Janssen J. & Higgs D. (2007) Attraction and localization of round goby (*Neogobius melanostomus*) to conspecific calls. *Behaviour*, 144, 1-21.

Yavno S. & Corkum L.D. (2009) Reproductive female round gobies (*Neogobius melanostomus*) are attracted to visual male models at a nest rather than to olfactory stimuli in urine of reproductive males. *Behaviour*, 147, 121-132.

Yavno S. & Corkum L.D. (2011) Round goby *Neogobius melanostomus* attraction to conspecific and heterospecific egg odours. *Journal of Fish Biology*, 78, 1944-1953.

#### **4.2.10 Increasing carbon dioxide concentrations**

- No evidence was captured on the use of carbon dioxide for management of invasive Ponto-Caspian gobies.

##### **Background**

Addition of dry ice to rapidly deliver a high dose of carbon dioxide, which results in a rapid reduction in dissolved oxygen levels, can sedate some fish species, with minimal residual toxicity. This may therefore offer a tool enabling selective removal of invasive gobies from waterbodies. This is provided that sufficiently high carbon dioxide levels can be maintained, which may be difficult, particularly in natural water bodies.

Increasing carbon dioxide concentrations by either bubbling the pressurised gas directly into water or by the addition of sodium bicarbonate, has been used to sedate fishes during transport or to allow handling of large numbers of fishes, with minimal residual toxicity (Clearwater *et al.* 2008). Previous studies have found that exposure to sodium bicarbonate at a concentration of 142–642 mg/litre for five minutes can kill some fish species (Brooke *et al.* 1978).

Brooke H.E., Hollender, B. & Lutterbie G. (1978) Sodium bicarbonate, an inexpensive fish anesthetic for field use. *The Progressive Fish-Culturist*, 40, 11–13.

Clearwater S.J., Hickey C.W. & Martin M.L. (2008) Overview of potential piscicides and molluscicides for controlling aquatic pest species in New Zealand. *Science for Conservation*, 283, 1-74.

#### **4.2.11 UV radiation**

- No evidence was captured on the use of UV radiation to control goby populations.

##### **Background**

Applying UV radiation to an invaded waterbody may offer a tool for localised eradication or population reduction of gobies, provided potentially negative effects on native species are carefully managed. A combined laboratory and field study in Lake Tahoe, USA indicated that UV levels in parts of the lake are intense enough to kill all largemouth bass *Micropterus salmoides* larvae (Tucker *et al.* 2012).

Tucker A.J., Williamson C.E. & Oris J.T. (2012) Development and application of a UV attainment threshold for the prevention of warm water aquatic invasive species. *Biological Invasions*, 14, 2331-2342.

#### **4.2.12 Changing salinity**

- A replicated, controlled laboratory study in Canada<sup>1</sup> found 100% mortality of round gobies within 48 hours of exposure to water of 30% salinity.

##### **Background**

Changing the salinity of an invaded waterbody may offer a tool for localised eradication or population reduction of gobies, provided potentially negative effects on native species are managed carefully. Increasing the salinity to a very high level is likely to have significant effects on non-target species.

A replicated, controlled laboratory study from 2006 to 2007 at the Great Lakes Institute for Environmental Research in Canada<sup>1</sup> found that round gobies *Neogobius melanostomus* cannot survive for more than two days in water with 30% salinity. All fish survived five hours in water of 30% salinity. It did not make a difference if the water became salty gradually or immediately. Up to about a fifth of the fish were still alive after 24 hours. However, after 48 hours, all fish were dead. Gobies were taken from a river in Canada. Ten gobies were put in each of 12 aquaria containing 16 litres of filtered river water. The water in four of the aquaria had 30% salinity from the beginning. The salinity in another four aquaria was 4% at the start of the experiment and increased every hour to 8, 14, 24 and 30%. Every hour for five hours, and after 24 and 48 hours, dead gobies were removed and counted.

(1)Ellis S. & McIsaac H.J. (2009) Salinity tolerance of Great Lakes invaders. *Freshwater Biology*, 54, 77-89.

#### **4.2.13 Changing pH**

- No evidence was captured on the use of pH alteration to control goby populations.

##### **Background**

Some species of fish are sensitive to pH changes and this can offer a tool for localised eradication or population reduction, provided potentially negative effects on native species are carefully managed. Draining ponds and altering the pH has been shown to be effective in local eradication of other fish species. For example, a before-and-

after study in the UK found that draining ponds and altering pH using lime slurry, eradicated topmouth gudgeon *Pseudorasbora parva* (Britton *et al.* 2008).

Britton R., Brazier M., Davies G.D. & Chare S.I. (2008) Case studies on eradicating the Asiatic cyprinid *Pseudorasbora parva* from fishing lakes in England to prevent their riverine dispersal. *Aquatic Conservation: Marine and Freshwater Ecosystems*, 18, 867-876.

#### 4.2.14 Use of barriers to prevent migration

- A controlled, replicated field study in the USA<sup>1</sup>, found that an electrical barrier prevented movement of round gobies across it, and that increasing electrical pulse duration and voltage increased effectiveness of the barrier.

#### Background

A number of different systems can be used to prevent dispersal of fish to new habitats, or to prevent their migration. For example, a replicated study from the USA found that leaf litter could act as a barrier, preventing dispersal of the eastern mosquitofish *Gambusia holbrooki* (Alemadi & Jenkins 2008). Another controlled study from the USA found that sound bubble barriers prevented Asian carp (*Cyprinidae*) from moving upstream (Ruebush *et al.* 2012). Other systems include altered flow rates. However, one study concluded that water flow rates faster than 125 cm/second, along with a route free from rest and refuge areas, would be necessary to prevent round goby *Neogobius melanostomus* migrating upstream (Tierny *et al.* 2011). This was because round gobies were found to be able to hold their position in strong water currents for extended periods using their pectoral fins as brakes, and could recover rapidly from exhaustive exercise, achieving powerful speed bursts (Tierny *et al.* 2011). Other systems that can be used to prevent fish migration include electrical barriers.

Alemadi S.D. & Jenkins D.G. (2008) Behavioral constraints for the spread of the eastern mosquitofish, *Gambusia holbrooki* (*Poeciliidae*). *Biological Invasions*, 10, 59-66.

Ruebush B.C., Sass G.G., Chick J.H. & Stafford J.D. (2012) In-situ tests of sound-bubble-strobe light barrier technologies to prevent range expansions of Asian carp. *Aquatic Invasions*, 7, 37-48.

Tierny K.B., Kasurak A.V., Zielinski B.S. & Higgs D.M. (2011) Swimming performance and invasion potential of the round goby. *Environmental Biology of Fishes*, 92, 491-502.

A controlled, replicated field study in the Shiawassee River, Michigan, USA<sup>1</sup>, found that an electrical barrier prevented round goby *Neogobius melanostomus* movement across it. Without any electrical current, round goby crossed the barrier within 20 minutes from release upstream. Using electrical settings shown to inhibit passage in the laboratory, the only marked round goby found below the barrier were dead. At reduced pulse durations, a few round goby (on average one per test) were found alive, but debilitated, below the barrier. Increasing electrical pulse duration and voltage increased the effectiveness of the barrier. Feasibility studies in a 2 m donut-shaped tank determined the required electrical currents. In field studies, an electrical barrier was placed between two blocking nets. The barrier consisted of 6 m

wide canvas on which were laid four cables carrying the electrical current. Twenty five latex paint-marked round goby were introduced upstream of the electrical barrier and recovered 24 h later upstream, on or downstream of the barrier.

(1)Savino J.F., Jude D.J & Kostich M.J. (2001) Use of electric barriers to deter movement of round goby. *American Fisheries Society Symposium*, 26, 171-182

#### **4.2.15 Public education**

- No evidence was captured on the impact of education programmes on control of goby populations.

#### **Background**

Public education programmes on the risks of invasive gobies and the mechanisms of spread could reduce the translocation of gobies into new water bodies, thereby reducing population spread.

## 5 Invasive reptiles

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### 5.1 Red-eared terrapin *Trachemys scripta*

#### Background

The red-eared terrapin *Trachemys scripta* is a 20-60 cm freshwater turtle characterised by prominent yellow to red patches on each side of the head, typically red on *T. scripta elegans*, the most commonly traded subspecies of the 15 described (Scalera 2006). The red-eared terrapin is native to eastern USA and adjacent areas of northeastern Mexico (Scalera 2006). It has been introduced to many non-indigenous localities worldwide, primarily through the pet trade, and is now widely spread throughout Europe. The red-eared terrapin is able to tolerate a wide range of permanent water bodies, ranging from brackish waters to manmade canals, and urban ponds. It may be found throughout the UK, but most records of persistent individuals are from warmer or more southerly areas (Wilkinson 2012).

Successful reproduction of the red-eared terrapin has not been recorded in the UK as 59-112 consecutive days of warm weather are necessary for the eggs to hatch (Scalera 2006), but has been recorded in areas with warmer climates, such as southern Spain (Perez-Santigosa *et al.* 2008).

Red-eared terrapins feed on a wide variety of native species. Juvenile red-eared terrapins are highly carnivorous, whereas adults are omnivorous generalists that opportunistically consume aquatic invertebrates such as insects and molluscs, in addition to fish, frog eggs, tadpoles, aquatic snakes, and a wide variety of plants and algae (Ernst *et al.* 1994). The red-eared terrapin can also feed on bottom-rooting plants which may result in systems becoming more turbid.

To raise body temperature, the red-eared terrapin exhibits basking behaviour. In the UK, where the climate is relatively cool in comparison with its native range, basking time is relatively long. As a result, the red-eared terrapin is easily seen. The basking behaviour of red-eared terrapins has been suggested as a potential problem for nesting water birds such as moorhens *Gallinula chloropus* and coots *Fulica atra*, as turtles clambering onto nests can partially submerge these fragile nests, killing the eggs and chicks. The red-eared terrapin can threaten and outcompete native semi-aquatic turtles, such as the endangered (non-UK) European pond terrapin *Emys orbicularis* (CABI 2014).

Red-eared terrapins have been known to live up to 42 years in the wild, although most probably do not survive beyond 30 years (Harding 1997). It is a possibility that global climate change could enable successful breeding and population establishment in the UK, however, since EU wildlife trade regulations banned European import of the red-eared terrapin in 1997, temperatures would have to rise significantly within a relatively short time period for this invasive to present an ongoing risk.

CABI (2014) Datasheet on *Trachemys scripta elegans* (red-eared slider). CABI Invasive Species Compendium. 19pp.

Ernst C.H., Lovich J.E. & Barbour R.W. (1994) *Turtles of the United States and Canada*. Smithsonian Institution Press, Washington D.C., 578 pp.

Harding J.H. (1997) *Amphibians and Reptiles of the Great Lakes Region*. University of Michigan Press, Michigan, 400 pp.

Perez-Santigosa N., Diaz-Paniagua C. & Hildigo-Vila J. (2008) The reproductive ecology of exotic *Trachemys scripta elegans* in an invaded area of southern Europe. *Aquatic Conservation: Marine and Freshwater Ecosystems*, 18, 1302 – 1310.

Scalera R. (2006) Fact sheet on *Trachemys scripta*. Delivering Alien Invasive Species Inventories for Europe (DAISIE). 4pp.

Wilkinson J. (2012) Red-eared Terrapin, *Trachemys scripta*. GB Non-Native Species Secretariat. Sand Hutton, UK. 3pp.

## Key messages

### ***Direct removal of adults***

Two studies, a replicated study from Spain using Aranzadi turtle traps, and an un-replicated study in the British Virgin Islands using sein netting, successfully captured but did not eradicate red-eared terrapin populations.

### ***Biological control using native predators***

No evidence was captured on the potential to use native predators to reduce red-eared terrapin populations.

### ***Draining invaded waterbodies***

No evidence was captured on the impact of draining invaded waterbodies on reduction of red-eared terrapin populations.

### ***Search and removal using sniffer dogs***

No evidence was captured on the success of use of sniffer dogs in removing red-eared terrapins.

### ***Application of a biocide***

A replicated, controlled laboratory study in the USA, found that application of glyphosate to the eggs of red-eared terrapins reduced hatching success to 73% but only at the highest experimental concentration of glyphosate and a surface active agent.

### ***Public education***

No evidence was captured on the impact of education programmes on reduction of red-eared terrapin populations.

### **5.1.1 Direct removal of adults**

- A replicated field study in Spain<sup>1</sup> found that Aranzadi turtle traps were effective in trapping red-eared terrapins from a river but did not eradicate the population.
- A study in the British Virgin Islands<sup>2</sup> found that using sein nets to trap adults and juveniles was not successful in eradicating the population.

## Background

Direct removal of adults by trapping, netting, shooting, or hand capture, may offer a tool for localised eradication, particularly in England, UK where there are no known instances of successful reproduction in the wild.

A variety of traps are referenced in the literature. For example, a fact sheet on the red-eared terrapin *Trachemys scripta* references successful use of floating boards with baited cages on top by terrapins as basking sites. Trap preference is sometimes determined by the level of visibility to the public in addition to efficacy (Bringsøe 2006). An information bulletin references that in Australia, funnelled 'Cathedral traps' are used in preference to 'basking traps' which are difficult to transport and unsuitable for use in public or high visibility locations (O'Keefe 2009). Some studies have researched the impact of bait location on trap success. For example, a replicated field study in the USA researched bait location and found no significant difference in trapping rate between traps with bait suspended near the funnel entrance, and traps with bait filled containers (Nall & Thomas 2009).

No studies have been found that reference successful local eradication using trapping or netting techniques.

Bringsøe H. (2006) *Trachemys scripta*. NOBANIS – Invasive Alien Species Fact Sheet. European Network on Invasive Alien Species, 13pp.

Nall I. & Thomas R. (2009). Does method of bait presentation within funnel traps influence capture rates of semi-aquatic turtles? *Herpetological Conservation and Biology* 42, 161-163.

O'Keefe S. (2009) The Practicalities of Eradicating Red-eared Slider Turtles (*Trachemys scripta elegans*). *Aliens: The Invasive Species Bulletin. Newsletter of the IUCN/SSC Invasive Species Specialist Group*. 28, 19-24.

A replicated field study conducted in 2008, in the Arga River, Spain<sup>1</sup> found that modified Aranzadi turtle traps were effective at trapping red-eared terrapin *Trachemys scripta elegans* and *Trachemys scripta scripta* but did not eradicate the populations, and that these traps performed better than modified Bolue traps, and fish-baited traps, which trapped very few terrapins. The modified Aranzadi turtle traps caught an average of 70% of observed terrapins. During five months of spring and summer 2008, one of each of the three trap types was set in each of two areas of the Arga River, Spain. On separate dates, one of each trap type was also set in 11 different water bodies. The baited traps were visited on consecutive days, while basking traps were checked weekly during five months of spring and summer of 2008.

A study in 2003 in a pond at botanic gardens in Tortola, British Virgin Islands<sup>2</sup> found that using sein nets to trap red-eared terrapin *Trachemys scripta* adults and juveniles was not successful in eradicating the population. Twelve adults and approximately twenty juveniles were removed. Additional capture efforts removed further adults and juveniles in July and October 2004. Experimental methods were not available.

(1) Valdeón A., Crespo-Diaz A., Egaña-Callejo A. & Gosá A. (2010) Update of the pond slider *Trachemys scripta* (Schoepff, 1792) records in Navarre (Northern Spain), and presentation of the Aranzadi Turtle Trap for its population control. *Aquatic Invasions*, 5, 297-302.

(2) Perry G., Owen J.L., Petrovic C., Lazell J. & Egelhoff J. (2007) The red-eared slider, *Trachemys scripta elegans*, in the British Virgin Islands. *Applied Herpetology*, 4, 88-89.

### **5.1.2 Biological control using native predators**

- No evidence was captured on the use of predators to control invasive terrapin populations.

#### **Background**

A study in the USA provided a strong indication that racoons feed on adult red-eared terrapins *Trachemys scripta* in their native range, with female terrapins smaller than 200mm in plastron length at particularly high risk (Tucker *et al.* 1999).

Although hatchlings of red-eared terrapins could conceivably be eaten by a few native UK species including otters (e.g. European otter *Lutra lutra*), herons (e.g. grey heron *Ardea cinerea*), rodents or corvids, or even introduced species like the American mink *Neovison vison*, once adult this species would have few natural enemies in the UK (Bringsøe 2006, Wilkinson 2012).

Successful reproduction of red-eared terrapins has not been recorded in the UK (Scalera 2006). As native UK predators are unlikely to be large enough to eat adult terrapins it is unlikely that population control using natural predators could deliver a control mechanism for UK populations.

Bringsøe H. (2006) *Trachemys scripta*. NOBANIS – Invasive Alien Species Fact Sheet. European Network on Invasive Alien Species, 13pp.

Scalera R. (2006) Fact sheet on *Trachemys scripta*. Delivering Alien Invasive Species Inventories for Europe (DAISIE). 4pp.

Tucker J.K., Filoramo N.I. & Janzen F.J. (1999) Size-based mortality due to predation in a nesting freshwater turtle, *Trachemys scripta*. *American Midland Naturalist*, 141, 198-203.

Wilkinson J. (2012) Red-eared Terrapin, *Trachemys scripta*. GB Non-Native Species Secretariat. Sand Hutton, UK. 3pp.

### **5.1.3 Draining invaded waterbodies**

- No evidence was captured on the impact of draining invaded waterbodies on reduction of red-eared terrapin populations.

## Background

Draining invaded waterbodies may offer a tool for localised management of red-eared terrapin *Trachemys scripta* populations, provided potentially negative side effects on native species are carefully managed. Following draining, red eared terrapins may burrow into the silt at the bottom of the waterbody. For example, a field trial in Queensland, Australia drained a small irrigation dam and found that red-eared terrapins burrowed to a depth of up to 2 m in the silt at the bottom and had to be removed by mechanical excavator (O’Keefe 2009). The silt was spread in a secure area, raked, and any red-eared terrapins present were removed by hand. To prevent red-eared terrapins from emigrating during the draining process, the site was first secured with barrier fences and pitfall traps. No additional details about the methods or results were provided.

O’Keefe S. (2009) The Practicalities of Eradicating Red-eared Slider Turtles (*Trachemys scripta elegans*). *Aliens: The Invasive Species Bulletin. Newsletter of the IUCN/SSC Invasive Species Specialist Group.* 28, 19-24.

### 5.1.4 Search and removal using sniffer dogs

- No evidence was captured on the success of use of sniffer dogs in removing red-eared terrapins.

## Background

Sniffer dogs can reportedly be used to detect and remove both red-eared terrapins *Trachemys scripta* and their eggs (Bringsøe 2006). Therefore, it is possible that sniffer dogs can be used to reduce or eradicate local invasive populations.

Bringsøe H. (2006) *Trachemys scripta*. NOBANIS – Invasive Alien Species Fact Sheet. European Network on Invasive Alien Species, 13pp.

### 5.1.5 Application of a biocide

- One replicated, controlled laboratory study in the USA<sup>1</sup>, found that application of glyphosate to the eggs of red-eared terrapins reduced hatching success to 73%, but only at the highest experimental concentration of glyphosate and a surface active agent.

## Background

Biocides such as glyphosate may negatively impact red-eared terrapin *Trachemys scripta* populations, by reducing hatching success of the eggs of breeding populations.

A replicated, controlled laboratory study in 2005 in the USA<sup>1</sup> found that application of glyphosate to the eggs of red-eared terrapins *Trachemys scripta elegans* reduced hatching success and the health of hatchlings, but only at the highest glyphosate concentration. Hatching success at the highest concentration of 11,206 ppm wet weight of glyphosate in Glypro and 678 ppm of the surface active agent LI700 was 73%, compared to hatching success of 80-100% in the lower concentrations and the control. Hatchlings from eggs that had been exposed to the highest concentration of glyphosate and surface active agent also weighed less both at hatching and at the end of the holding period, compared to those from eggs that had been exposed to lower concentrations. Eggs of red-eared terrapins were exposed to single applications of glyphosate and surface active agent, ranging from 0 to 11,206 ppm wet weight of glyphosate in Glypro and 0 to 678 ppm of the surface active agent LI700.

(1) Sparling D., Matson C., Bickham J. & Doelling-Brown P. (2006) Toxicity of glyphosate as glypro\_ and LI700 to red-eared slider (*Trachemys scripta elegans*) embryos and early hatchlings. *Environmental Toxicology and Chemistry*, 25, 2768–2774.

### **5.1.6 Public education**

- No evidence was captured on the impact of education programmes on reduction of red-eared terrapin populations.

#### **Background**

Educating the public on the negative effects of releasing red eared terrapins *Trachemys scripta* from captivity could help to reduce the number of terrapins in the wild (Bringsøe 2006).

Bringsøe H. (2006) *Trachemys scripta*. NOBANIS – Invasive Alien Species Fact Sheet. European Network on Invasive Alien Species, 13pp.

## 6 Invasive amphibians

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### 6.1 The American bullfrog *Lithobates catesbeiana*

#### Background

The American Bullfrog *Lithobates catesbeiana* is a widely introduced and invasive anuran that is frequently blamed for population declines of indigenous species (Adams & Pearl 2007). Introduced bullfrogs can have devastating effects on wildlife that evolved without equivalent predatory types. The American bullfrog has been blamed for amphibian declines in much of western North America, and potentially serves as a vector of diseases to native amphibians (Fisher & Garner 2007). They can inhabit most permanent water sources including canals, reservoirs, marshes, ponds, and lakes and the tadpoles require only perennial water and grazeable plant material to survive and grow (Rosen & Schwalbe 1995). Bullfrogs can live at extremely high densities, and when densities are reduced (such as after an unsuccessful eradication attempt), their survival and successful reproductive rates increase resulting in a rapid population rebound. Bullfrogs can travel distances over land of up to 1 km to colonize new water sources (Miera 1999).

Adams M.J. & Pearl C.A. (2007) Problems and opportunities managing invasive bullfrogs: is there any hope? Pages 679–693 in: F. Gherardi (eds) *Biological invaders in inland waters: profiles, distribution and threats*, Springer, Dordrecht, The Netherlands.

Fisher M.C & Garner T.W.J (2007) The relationship between the emergence of *Batrachochytrium dendrobatidis*, the international trade in amphibians and introduced amphibian species. *Fungal Biology Reviews*, 21, 2-9.

Rosen, P.C. & Schwalbe C.R. (1995). Bullfrogs: introduced predators in southwestern wetlands. Pages 452-454 in: E. T. LaRoe, G. S. Farris, C. E. Puckett, P. D. Doran & M. J. Mac (eds) *Our living resources: a report to the nation on the distribution, abundance, and health of US plants, animals, and ecosystems*. U.S. Department of the Interior, Washington D.C., USA.

Miera V. (1999) Simple introductions – major repercussions: The story of bullfrogs and crayfish in Arizona. *Arizona Wild Views*, 42, 25-27.

#### Key messages

##### ***Biological control using native predators***

One replicated, controlled study conducted in north-eastern Belgium, found the introduction of the northern pike led to a strong decline in bullfrog tadpole numbers.

##### ***Biological control of co-occurring beneficial species***

No evidence was captured on the effects of removing co-occurring beneficial species on the control of American bullfrogs.

##### ***Habitat modification***

No evidence was captured on the effects of habitat modification on the control of American bullfrogs.

##### ***Draining ponds and altering the length of time for which the pond contains water***

No evidence was captured on the effects of draining ponds and altering the length of time for which the pond contains water on the control of American bullfrogs.

### ***Pond destruction***

No evidence was captured on the effects of pond destruction on the control of American bullfrogs.

### ***Fencing***

No evidence was captured on the effects of fencing on the control of American bullfrogs.

### ***Direct removal of adults***

One replicated study in Belgium found catchability of adult bullfrogs in small shallow ponds using one double fyke net to be very low. One small study in the USA, found that bullfrog adults can be captured overnight in a single trap floating on the water surface. One replicated, controlled study in the USA found that bullfrog populations rapidly rebounded following intensive removal of the adults. One study in France found a significant reduction in the number of recorded adults and juveniles following the shooting of metamorphosed individuals before reproduction, when carried out as part of a combination treatment.

### ***Direct removal of juveniles***

One replicated study in Belgium found double fyke nets were effective in catching bullfrog tadpoles in small shallow ponds. One study in France found a significant reduction in the number of recorded adults and juveniles following the removal of juveniles by trapping, when carried out as part of a combination treatment.

### ***Collection of egg clutches***

No evidence was captured on the effects of collection of egg clutches on the control of American bullfrogs.

### ***Application of a biocide***

One replicated, controlled study in the USA found a number of chemicals killed American bullfrogs, including caffeine (10% solution), chloroxylenol (5% solution), and a combined treatment of Permethrin (4.6% solution) and Rotenone (1% solution).

### ***Public education***

No evidence was captured on the effects of public education programmes on the control of American bullfrogs.

## **6.1.1 Biological control using native predators**

- One replicated, controlled study conducted in Belgium<sup>1</sup>, found the introduction of the northern pike led to a strong decline in bullfrog tadpole numbers.

### **Background**

Introduction of native predators can increase predation on bullfrog tadpoles, thereby biologically controlling bullfrog populations.

One replicated, controlled study conducted from 2007 to 2009 in Balen, north-eastern Belgium<sup>1</sup>, found the introduction of the northern pike *Esox lucius* led to a

reduction in bullfrog tadpole biomass with time, which was not significant overall, but highly significant from Spring year two. In year two, tadpole biomass in ponds with introduced pike reached only a tenth of their biomass in control (unmanaged) treatments in year two. No effect of draining was observed. Four treatments were randomly assigned to twelve ponds. The control included two replicates with no draining and no pike. The second treatment included four replicates of pike, but no draining. The third included three replicates of draining and no introduction of pike. The fourth included three replicates of pike and draining. Draining was performed in June 2007, with removal of all amphibians and fish. Juvenile pike were introduced in May 2008 and 2009.

(1) Louette G. (2012). Use of a native predator for the control of an invasive amphibian. *Wildlife Research*, 39, 271-278.

### **6.1.2 Biological control of co-occurring beneficial species**

- No evidence was captured on the effects of removing co-occurring beneficial species on the control of American bullfrogs.

#### **Background**

American bullfrog populations can benefit from the presence of other species, such as invasive fishes. Reducing the population of co-occurring beneficial species in localised populations, and limiting their spread or intentional introduction, may offer a tool for managing and reducing bullfrog populations. For example, one replicated, controlled field experiment in Oregon, USA found that the invasive bluegill sunfish increased the survival rate of bullfrog tadpoles by reducing the abundance of native aeshnid dragonfly nymphs (Adams *et al.* 2003). Treatments consisted of either one bluegill or no fish plus either twelve recently hatched aeshnid dragon fly nymphs and three aeshnid dragonfly nymphs close to metamorphosis, or no nymph). Fifty bullfrog tadpoles were added to each enclosure. This was a three year study during which 85 ponds and wetlands were surveyed.

Adams M. J., Pearl C. A., & Bury R. B. (2003). Indirect facilitation of an anuran invasion by non-native fishes. *Ecology Letters* 6, 343–351.

### **6.1.3 Habitat modification**

- No evidence was captured on the effects of habitat modification on the control of American bullfrogs.

## Background

Habitat modification such as increasing shade for native species, removing protective cover, or breaking up wet travel corridors may offer a technique to indirectly reduce bullfrog populations and establish or increase native amphibian populations. For example, bullfrogs have been found to be less abundant in ponds with shallow sloping banks and extensive emergent vegetation (Adams *et al.* 2003).

Adams M. J., Pearl C. A., & Bury R. B. (2003). Indirect facilitation of an anuran invasion by non-native fishes. *Ecology Letters*, 6, 343–351.

### **6.1.4 Draining ponds and altering the length of time for which the pond contains water**

- No evidence was captured on the effects of draining ponds or altering the length of time for which ponds contain water on the control of American bullfrogs.

## Background

Breeding bullfrog populations have been found to disappear following natural pond drying (Maret *et al.* 2006). Therefore, draining invaded waterbodies may offer a tool for localised eradication or population reduction of American bullfrogs, provided potentially negative effects on native species are carefully managed (Maret *et al.* 2006). It is also possible to prevent bullfrog larvae from completing metamorphosis by selective draining to reduce the length of time for which the pond contains water (Govindarajulu 2004).

Maret T. J., Snyder J. D. & Collins J. P. (2006). Altered drying regime controls distribution of endangered salamanders and introduced predators. *Biological Conservation*, 127, 129-138.

Govindarajulu P. (2004) Introduced bullfrogs (*Rana catesbeiana*) in British Columbia: impacts on native Pacific treefrogs (*Hyla regilla*) and red-legged frogs (*Rana aurora*). *PhD thesis. University of Victoria, Victoria.*

### **6.1.5 Pond destruction**

- No evidence was captured on the effects of pond destruction on the control of American bullfrogs.

## Background

Pond destruction is a potential tool for removing the breeding site, and can be useful provided that bullfrogs are prevented from dispersing to new sites, and provided that there are no significant impacts on non-target organisms.

### **6.1.6 Fencing**

- No evidence was captured on the effects of fencing on the control of American bullfrogs.

#### **Background**

Fencing can be used to reduce population dispersal, thereby reducing the spread of the American bullfrog to neighbouring water bodies. This is particularly useful to contain new populations, and also for use during other control efforts.

### **6.1.7 Direct removal of adults**

- One replicated study in Belgium<sup>1</sup> found catchability of adult bullfrogs in small shallow ponds using one double fyke net for 24 h to be very low.
- One small study in the USA<sup>2</sup>, found that bullfrog adults can be captured overnight in a single trap floating on the water surface.
- One replicated, controlled study in the USA<sup>3</sup> found that bullfrog populations rapidly rebounded following intensive removal of the adults.
- One before-and-after study in France<sup>4</sup> found a significant reduction in the number of recorded adults and juveniles following the shooting of metamorphosed individuals before reproduction, when carried out as part of a combination treatment.

#### **Background**

Direct removal of adults by trapping, shooting, hand spearing, hand capture, use of artificial refuges, or electro-shocking, may offer a tool for localised population reduction when used as part of an integrated pest management strategy for controlling invasive bullfrog populations.

One replicated study in 2012 and 2013 in Balen, northeast Belgium<sup>1</sup>, found catchability of adult bullfrog in small shallow ponds using one double fyke net for 24 h to be reasonably consistent at approximately 0.7%. Catchability of adult American bullfrogs was investigated using mark-recapture at the peak of reproduction. Adult bullfrogs were sampled during 16 separate capture occasions. At each sampling occasion in 2012, ten ponds were randomly sampled, and six ponds at each capture occasion in 2013. For every subsequent sampling occasion, a new randomisation of sampled ponds was made. Each time, one double fyke net was placed 2m out and parallel to the shore of the longest side of the pond for 24 h. Every two days, fyke nets were alternated between the opposite banks of the ponds under investigation.

Individuals were marked with an injection of pigment under the skin, and released in the centre of the pond.

One small study in 2008 and 2009 in Colorado, USA<sup>2</sup>, found that bullfrog adults were captured overnight in a single trap floating on the water surface, but that shoreline trapping was relatively unsuccessful. Shoreline trapping only captured one bullfrog across two ponds, each with two traps, in 10 total trap nights in 2008. However, two floating traps placed in a third pond in 2009 captured 18 bullfrogs in 10 trap nights. Thirteen additional bullfrogs were removed by hand netting. All attractants trialled captured bullfrogs, and the rate of capture did not differ among types. In all ponds, two traps were tested, each 69 × 69 × 25 cm and constructed with 1.3 × 1.3 cm wire mesh. In 2009, the traps were modified so they floated by attaching Styrofoam flotation devices to the underside of the traps. A range of attractants were added to the traps, including lights, live crickets, and fishing lures, with various combinations of attractants tested for one to four nights.

One replicated, controlled study from 1986-1989 and 1992-1993, in the San Bernardino National Wildlife Refuge, Cochis County, Arizona, USA<sup>3</sup>, found that bullfrog populations rapidly rebounded following intensive removal of the adults. At one study pond, 854 large (80+ cm body length) bullfrogs had been removed from about 0.2 ha of habitat. After three to four active-season months, a 50-80% rebound toward pre-removal numbers was observed, together with weak evidence of positive effects on native leopard frogs and garter snakes. From 1986-1989 and 1992-1993, intensive bullfrog removals were conducted two to three times per year using funnel traps, hand spears, guns, and hand capture. Simultaneous monitoring of native ChiriCahua leopard frogs and Mexican garter snakes was carried out at the sites of bullfrog removal.

One before-and-after study from 2006 to 2009 on Natural Park Périgord-Limousin sites, France<sup>4</sup> reported a significant reduction in the number of recorded adults and juveniles following the shooting of metamorphosed individuals before reproduction, along with trapping of juveniles and collection of egg clutches. The number of sighted and destroyed bullfrogs decreased from 130-140 in 2006 and 2007, to approximately 80 in 2008, and fewer than 40 in 2009. It was reported that most of the bullfrogs eradicated by shooting were males due to ease of location relative to the silent females. Shooting with airguns was carried out at night using two person teams. One person was responsible for tracking and identification, and the other was responsible for eradication.

(1) Louette G., Devisscher S. & Adriaens T. (2014). Combatting adult invasive American bullfrog *Lithobates catesbeianus*. *European Journal of Wildlife Research*, 60, 703–706.

(2) Snow N.P. & Witmer G. W. (2011). A field evaluation of a trap for invasive American bullfrogs. *Pacific Conservation Biology*, 17, 285-291.

(3) Rosen, P.C. & Schwalbe C.R. (1995). Bullfrogs: introduced predators in southwestern wetlands. Pages 452–454 In: E. T. LaRoe, G. S. Farris, C. E. Puckett, P. D. Doran & M. J. Mac (eds) *Our living resources: a report to the nation on the distribution, abundance, and health of US plants, animals, and ecosystems*. U.S. Department of the Interior, Wasington D.C., USA.

(4) Guibert S., Dejean T. & Hippolyte S. (2010). Le Parc naturel régional Périgord-Limousin: territoire d'expérimentation et d'innovation par la mise en place d'un programme d'éradication de la Grenouille taureau (*Lithobates catesbeianus*) associé à un programme de recherche sur les maladies émergentes des amphibiens. *EPOPS*, 79, 15-24.

### 6.1.8 Direct removal of juveniles

- One replicated study in Belgium<sup>1</sup> found double fyke nets were effective in catching bullfrog tadpoles in small shallow ponds.
- One before-and-after study in France<sup>2</sup> found a significant reduction in the number of recorded adults and juveniles following the removal of juveniles by trapping, when carried out as part of a combination treatment.

#### Background

Direct removal of the early lifecycle stage may offer a tool for localised population reduction when used as part of an integrated pest management strategy for controlling invasive bullfrog populations. For example, a replicated field based and modelling study from 1999 to 2003 on Southern Vancouver Island, Canada (Govindarajulu *et al.* 2005) found that culling bullfrog metamorphs in autumn was the most effective method of decreasing population growth rate.

Govindarajulu P., Altwegg R. & Anholt B.R. (2005) Matrix model investigation of invasive species control: bullfrogs on Vancouver Island. *Ecological Applications*, 15, 2161–2170.

A replicated study in 2010 and 2011 across three sites in Belgium<sup>1</sup> found catchability of bullfrog tadpoles in small shallow ponds using one double fyke net for 24 h to be reasonably consistent at approximately 6%. Bullfrog populations were investigated in ten permanently wet, small, shallow fish ponds (average surface area 1,500 m<sup>2</sup>; max depth 150 cm), across three sites. In six water bodies (Hoogstraten and Arendonk), bullfrog tadpole population density was estimated. In these ponds, a number of double fyke nets were set (parallel and two meters out from the shore) for 24 h, covering all sides of the water body. A minimum of three catch efforts of equal magnitude were performed. After every catch effort, all captured individuals were removed from the population. To determine the accuracy of these population size estimates, calibration using seine netting was performed in two ponds.

A before-and-after study from 2006 to 2009 on Natural Park Périgord-Limousin sites in France<sup>2</sup> found a significant reduction in the number of recorded adults and juveniles following the removal of juveniles by trapping, along with other removal methods. The number of trapped tadpoles decreased from approximately 1,600 in 2006 to fewer than 200 in 2009. Trapping was carried out as part of a combination treatment which also involved shooting of adults and collection of egg clutches. Unbaited single and double entry traps were installed equidistant from each other in the water, and were checked daily until the catch rate became negligible compared to the work effort.

(1) Louette G., Devisscher S. & Adriaens T. (2014). Combatting adult invasive American bullfrog *Lithobates catesbeianus*. *European Journal of Wildlife Research*, 60, 703–706.

(2) Guibert S., Dejean T. & Hippolyte S. (2010). Le Parc naturel régional Périgord-Limousin: territoire d'expérimentation et d'innovation par la mise en place d'un programme d'éradication de la Grenouille taureau (*Lithobates catesbeianus*) associé à un programme de recherche sur les maladies émergentes des amphibiens. *EPOPS*, 79, 15-24.

### 6.1.9 Collection of egg clutches

- Despite reference to removal of egg clutches in some studies using bilge pumps or nets, no evidence was captured on the effects of egg collection on American bullfrogs.

#### Background

Depending on body size, a female bullfrog may deposit 1,000 to 40,000 eggs, which hatch in 3-5 days (Snow & Witmer 2010). Removal of egg clutches offers a potential tool for reducing bullfrog populations (Guibert *et al.* 2010). However, for bullfrogs and some other temperate frogs, incomplete removal of eggs or larvae can boost growth and survival of remaining individuals via strong density dependence (Adams & Pearl 2007).

Snow N.P. & Witmer G. (2010) American Bullfrogs as Invasive Species: A Review of the Introduction, Subsequent Problems, Management Options, and Future Directions. *Proceedings Of the 24th Vertebrate Pest Conference* (Timm, RM & Fagerstone, KA, Eds.) Published at University of California, Davis. 2010.

Guibert S., Dejean T. & Hippolyte S. (2010) Le Parc naturel regional Périgord-Limousin: territoire d'expérimentation et d'innovation par la mise en place d'un programme d'éradication de la Grenouille taureau (*Lithobates catesbeianus*) associé à un programme de recherche sur les maladies émergentes des amphibiens. *EPOPS*, 79, 15-24.

Adams M.J. & Pearl C.A. (2007) Problems and opportunities managing invasive bullfrogs: is there any hope? Pages 679–693 in: F. Gherardi (eds) *Biological invaders in inland waters: profiles, distribution and threats*, Springer, Dordrecht, The Netherlands.

### 6.1.10 Application of a biocide

- One replicated, controlled study in the USA<sup>1</sup> found a number of chemicals killed American bullfrogs, including caffeine (10% solution), chloroxylonol (5% solution), and a combined treatment of Permethrin (4.6% solution) and Rotenone (1% solution).

#### Background

Biocides may offer a tool for localised eradication or population reduction of American bullfrogs, provided potentially negative effects on native species are carefully managed.

A replicated, controlled laboratory study from 2008 to 2009 at the University of California, USA<sup>1</sup>, reported that a number of chemicals killed American bullfrog. Caffeine (10% solution), chloroxylonol (5% solution), and a combined treatment of Permethrin (4.6% solution) and Rotenone (1% solution) each achieved 100% mortality. Dosed on their own, Permethrin (4.6% solution) and Rotenone (1%

solution) each achieved 40% mortality. In the trial, approximately 4 ml of treatment solution was sprayed on the entire dorsal surface of randomly-selected groups of bullfrogs using a handheld plastic spray bottle. There were five bullfrogs in each group. Water was used as the solvent for all materials. To improve solubility, a small amount of sodium benzoate was added to the caffeine solution, and a small amount of alcohol was added to the chloroxyleneol solution.

(1) Snow N.P. & Witmer G. (2010) American Bullfrogs as Invasive Species: A Review of the Introduction, Subsequent Problems, Management Options, and Future Directions. *Proceedings of the 24th Vertebrate Pest Conference* (Timm, R.M. & Fagerstone, K.A. Eds.) Published at University of California, Davis.

### **6.1.11 Public education**

- No evidence was captured on the effects of public education on the control of American bullfrogs.

#### **Background**

Public education programs on the risks of invasive American bullfrogs and the mechanisms of spread could reduce the movement of eggs and tadpoles as bait into waterways or ponds, and reduce further dispersal.